

**EVALUATION OF FRANKFURTERS FORMULATED WITH POTASSIUM
LACTATE AND SODIUM DIACETATE AND INOCULATED WITH *LISTERIA*
MONOCYTOGENES BEFORE AND AFTER IRRADIATION TREATMENT**

A Dissertation

by

TIMOTHY DAVID KNIGHT

Submitted to the Office of Graduate Studies of
Texas A&M University
in partial fulfillment of the requirements for the degree of

DOCTOR OF PHILOSOPHY

May 2006

Major Subject: Food Science and Technology

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ABSTRACT

Evaluation of Frankfurters Formulated with Potassium Lactate and Sodium Diacetate and Inoculated with *Listeria monocytogenes* Before and After Irradiation Treatment.

(May 2006)

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Microbial safety and quality attributes were evaluated for frankfurters formulated with potassium lactate/sodium diacetate (0 or 3%) and inoculated with a four-strain *Listeria monocytogenes* cocktail before and after treatment with pasteurizing doses of irradiation (0, 1.8, or 2.6 kGy). Frankfurters were inoculated after irradiation and stored aerobically for 4 wk at 4 °C to simulate the product becoming contaminated after opening, or they were inoculated prior to vacuum packaging and stored for 8 wk at 4 °C.

Incorporation of lactate/diacetate into frankfurter formulations with or without irradiation had a strong listeristatic effect throughout 4 wk of aerobic storage. Total microbial counts for frankfurters formulated with lactate/diacetate remained constant throughout storage while those without increased steadily (5.4 to 9.3 log cfu). Over 4 wk of storage, the outgrowth of *L. monocytogenes* on frankfurters formulated with lactate/diacetate was effectively suppressed and counts were not significantly different from initial counts (5.2 vs. 5.0 log cfu, respectively). Irradiation treatments alone had significantly higher *L. monocytogenes* counts after 3 wk of storage. Both treatments

together or alone were not detrimental to sensory aroma or flavor attributes. Meaty/brothy complex, smoke, spice aroma, springiness, and cohesiveness attributes were judged slightly lower for frankfurters formulated without lactate/diacetate than those with lactate/diacetate at the end of aerobic storage. Sensory color was not dramatically influenced by either treatment, however, $L^*a^*b^*$ values of all treatments decreased slightly during storage.

Both the addition of lactate/diacetate to a frankfurter formulation and irradiation were effective towards controlling microbial growth of *L. monocytogenes* in an unopened vacuum package after 8 wk of storage. Large and incremental reductions in total microbial counts were seen with irradiation treatment, which were maintained throughout storage with lactate/diacetate treated frankfurters. There were fewer influences on sensory characteristics for vacuum packaged frankfurters compared to those aerobically packaged.

Overall, lactate/diacetate addition and irradiation to a lesser extent were effective towards retarding the outgrowth *L. monocytogenes* on frankfurters while maintaining quality attributes throughout aerobic storage. The combination of irradiation and lactate/diacetate were effective for reducing and retarding growth of *L. monocytogenes* and especially during the last two weeks of vacuum packaged storage.

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CHAPTER I

INTRODUCTION

Listeria monocytogenes is a food-borne pathogen of immediate concern to human health. The publicity surrounding outbreaks of the organism associated with ready-to-eat (RTE) processed meats and the severity of illness have led to various intervention strategies that can be applied by a food manufacturer to help preserve the safety of products and reduce the incidence of listeriosis. An intervention strategy that addresses consumer safety concerns is the use of multiple microbial hurdles, which provide compatible and complementary anti-listerial mechanisms for suppressing pathogen growth while preserving desirable physiochemical and sensory properties.

This research evaluated the microbial safety and quality attributes of frankfurters formulated with or without a 60% solution of potassium lactate/sodium diacetate added at a 3% level. These frankfurters were inoculated with a four strain *Listeria monocytogenes* cocktail before or after treatment with pasteurizing doses of irradiation targeted at 0.0, 1.8, or 2.6 kGy. Phase 1 of this project investigated the effect of inoculation (10^3 CFU/frankfurter) *after* treatment with irradiation and subsequent aerobic storage at 4 °C to simulate an open package in a consumer's refrigerator. Phase 2 examined the effects of inoculation (10^5 cfu/frankfurter) *prior* to irradiation and vacuum packaged storage at 4 °C to simulate contamination before packaging. Samples for both phases were subjected to microbial evaluation, chemical/physical analysis, and sensory

This dissertation follows the style of the Journal of Food Science.

evaluation. This study was designed to answer important questions about the effectiveness of combined lactate/diacetate and irradiation treatments for increasing the safety of RTE meat products while maintaining quality attributes.

Research Hypothesis

Application of pasteurizing doses of electron beam irradiation in combination with a potassium lactate/sodium diacetate ingredient reduces the survival of *Listeria monocytogenes* inoculated onto frankfurters before and after irradiation and during subsequent storage at 4 °C.

Research Objectives

The specific objectives of this research were to evaluate the microbiological safety and quality attributes of frankfurters, formulated with potassium lactate/sodium diacetate, treated with electron-beam irradiation and inoculated before or after irradiation with *Listeria monocytogenes*.

This was done by:

1. Determining the survival/growth of *Listeria monocytogenes* inoculated onto the surface of frankfurters formulated with potassium lactate/sodium diacetate before and after treatment of the product with pasteurizing doses of irradiation and during storage at 4 °C.
2. Evaluating the oxidative stability, pH, and sensory properties of irradiation pasteurized frankfurters containing potassium lactate/sodium diacetate.

CHAPTER II

LITERATURE REVIEW

Listeriosis acquired from the consumption of RTE meat or poultry products represents a serious public health concern because of high mortality rates associated with the illness. The potential economic loss due to outbreaks and recalls of RTE products associated with *Listeria monocytogenes* has been well publicized (AMI 2002; CDC 1999, 2002; Mead and others 1999). For example, according to the Centers for Disease Control and Prevention (2002), 53 people were affected by an outbreak in the northeastern United States associated with precooked turkey deli meat contaminated with *L. monocytogenes*. The aftermath of this outbreak culminated in 53 illnesses including eight deaths and three miscarriages, the recall of 27 million pounds of fresh and frozen RTE turkey and chicken, and a voluntary month-long plant shutdown. Because of the devastation caused by this incident, the prevention of future outbreaks is imperative for all RTE meat processors.

Microbial Intervention Strategies for RTE Meats

In recent years, much research has focused on decontaminating RTE products using non-thermal and thermal intervention strategies. Combining strategies that are effective can have a synergistic effect and may be more effective for preventing the outgrowth of pathogens such as *L. monocytogenes* (Bedie and others 2001; Glass and others 2002; Islam and others 2002; Kozempel and others 2000; Lucore and others 2000; Muriana and others 2002, Samelis and others 2001, 2002). In addition to improving

safety, quality attributes can be retained over a longer period of time. Formulation with lactate/diacetate or irradiation treatment of packaged products are both effective for inhibiting growth of *L. monocytogenes*. When used in combination, they may serve as more potent anti-listerial agents and effectively reduce the level of irradiation needed to destroy *L. monocytogenes* in a RTE product. After irradiation and distribution, there is a risk of contamination by the consumer when the product is opened and not consumed immediately. If contamination with a pathogen such as *L. monocytogenes* occurs, there is potential for growth at refrigeration temperatures and subsequent food-borne illness. An irradiated product provides favorable conditions for this growth since existing microflora are typically lower, thus favoring *L. monocytogenes* growth. Lactate and diacetate are powerful anti-listeria agents and may prevent or retard the growth of the organism while the product is in consumer hands.

In RTE products, the principal safety concern is post-processing contamination of the product by *L. monocytogenes*, which is a particular problem because of its ability to grow at refrigeration temperatures. Not surprisingly, various interventions used for decontamination of meat/poultry carcasses have been employed to kill listeria in these products. Treatments have included heat pasteurization and organic acid rinses (Bedie and others 2001; Glass and others 2002; Islam and others 2002; Juncher and others 2000; Kozempel and others 2000; Lucore and others 2000; Mermelstein 2001; Muriana and others 2002; Samelis and others 2001, 2002). Heat treatments are not always effective on packaged products because many of the surfaces (e.g., between frankfurters, sliced products) are not fully exposed to lethal heat levels. Antimicrobial ingredients

with GRAS status are allowed in RTE product formulations or may be used as external treatments (Bedie and others 2001; Glass and others 2002; Keeton and others 2002; Samelis and others 2002).

Lactate as an Antimicrobial for RTE Meats

Lactate used in its various forms alone or in combination with other chemical antimicrobial ingredients is an effective antilisterial strategy. The antimicrobial effectiveness of lactate has been shown to be synergistic in the presence of other antimicrobials, most notably sodium diacetate. All lactate percentages in this chapter are stated as a percentage addition of a 60% (w/w) commercially available solution [40% water, 56% potassium lactate, 4% sodium diacetate]. Samelis and others (2002) looked at the effect of a combination of sodium lactate and sodium diacetate incorporated into frankfurter formulations which were surface inoculated with *L. monocytogenes* (10^4 cfu) after cooking but before vacuum packaging. The addition of 3% sodium lactate alone inhibited outgrowth of the organism for 35-50 d at refrigeration temperatures. However, 3% sodium lactate in combination with 0.25% sodium diacetate was even more effective for inhibiting listerial outgrowth for over 120 d storage. Mbandi and Shelef (2001, 2002) demonstrated similar anti-listerial effects for a combination of 2.5% sodium lactate and 0.2% sodium diacetate in both sterile comminuted beef and beef bologna. At both 5 and 10 °C, this combination of antimicrobials was listeristatic throughout 30 d storage, showing a 4.5 log cfu difference between samples with lactate/diacetate and those without at the end of storage. In a similar study reported by Bedie and others (2001), sodium lactate (3 or 6%) and sodium diacetate (0.25 or 0.5%) were incorporated

into frankfurters that were subsequently inoculated with *L. monocytogenes* (10^4 cfu). Each treatment alone prevented outgrowth compared to non-inoculated controls without additives throughout 120 d of vacuum packaged refrigerated storage. Combinations of the antimicrobials were not included in the experimental treatments, however, 6% sodium lactate and 0.5% sodium diacetate were the most effective treatments. Three percent sodium lactate and 0.25% sodium diacetate alone inhibited growth for 50-70 d.

Several authors have evaluated the incorporation of lactate in combination with dipping frankfurters in various antimicrobial solutions. Nunez and others (2004a) investigated the effectiveness of potassium lactate in a frankfurter formulation at 3.3% in combination with dipping treatments (30 s) of an acidified calcium sulfate (ACS) solution (with lactic and propionic acid), 3.3% potassium lactate, or 3.4% lactic acid against *L. monocytogenes* growth. Acidified calcium sulfate, and to a lesser extent lactic acid, had a significant surface bactericidal effect through 12 wk of refrigerated vacuum packaged storage regardless of the inclusion or exclusion of lactate in the frankfurter formulation. Sensory and physiochemical changes of all treatments were minimal throughout refrigerated storage (Nunez and others 2004b). Barmpalia and others (2004) used a combination of 1.8% sodium lactate and 0.25% sodium diacetate in frankfurters with dip solutions of 2.5% lactic or acetic acids. All treatments with lactate/diacetate included in the formulation demonstrated inhibition of outgrowth during 40 d of refrigerated vacuum packaged storage. Dipping inoculated frankfurters without lactate/diacetate for 2 minutes in either solution reduced initial *L. monocytogenes* 0.7 to 2.1 log cfu, however, the microbial counts increased rapidly during storage with no

residual activity. Again, sensory and physiochemical properties of frankfurters were unaltered in minimally changed treatments. Uhart and others (2004) looked at the antimicrobial compound pediocin in combination with sodium lactate (6%) and sodium diacetate (3%) dips to control *L. monocytogenes* on vacuum packaged beef frankfurters during 3 wk refrigerated storage. The combination of lactate and diacetate decreased the population 3 log cfu units from 10^7 to 10^4 cfu after 3 wk storage. The addition of pediocin added an additional half log cfu reduction to the effect of lactate and diacetate at 3 wk.

A number of articles have been published which detail the influence of lactate incorporation into processed meats. In each case, lactate had a strong bacteriostatic influence during storage at levels around 3% in the final product. A series of papers detail the antimicrobial effectiveness and sensory properties of cooked uncured top beef rounds injected with lactate before processing. Papadopoulos and others (1991a) looked at the sensory properties of uncured cooked beef top rounds treated with sodium lactate at 3% and stored at 0°C for 84 d. Added lactate increased cook yields and APC counts were lower for lactate formulated roasts than for those without added lactate. During storage, *Lactobacillus* spp. became the predominant microflora. A trained panel evaluation determined that the desirable fresh beef flavor notes were more predominant for lactate-formulated roasts, which was preferred over those without lactate by consumer panels with flavor and texture driving higher acceptability. Few sensory changes occurred in any treatment over storage. Papadopoulos and others (1991b) looked at these same treatments and determined that the microbiological quality of the

lactate formulated vacuum packaged roasts was at least 84 d. Papadopoulos and others (1991c) also performed a more detailed descriptive attribute sensory analysis of the same treatments which showed retention of positive beef flavor attributes and a decrease in flavor attributes associated with warmed-over during storage for lactate formulated roasts compared to those without lactate. Sodium lactate did not adversely affect sensory attributes or quality of vacuum packaged cooked beef roasts. In addition, Miller and Acuff (1994) looked at the effectiveness of sodium lactate injected at 1, 2, 3, or 4% into beef top rounds against several pathogenic microorganisms inoculated onto the surface and stored at 10 °C for 28 d. Lactate was found to have a bacteriostatic effects at 3 and 4% concentrations against *L. monocytogenes*, *Salmonella typhimurium*, and *Escherichia coli* O157:H7. Maca and others (1999) looked at temperature effects for vacuum packaged cooked top rounds held at 0, 4, 10, or 16 °C for 3 wk for the same treatments. They showed that 4% sodium lactate was effective at controlling the microbial population even under temperature abuse situations. In addition, the same sensory quality trends seen with Papadopoulos' work occurred at higher storage temperatures as well. Maca and others (1997b) also looked at the combination of sodium lactate (3 or 4%) alone and in combination with sodium propionate (0.1 or 0.2%) for treating vacuum packaged cooked top rounds stored for 84 d at 4 °C. All treatment combinations had a bacteriostatic influence and enhanced positive beef flavor attributes.

The influence of lactate on ground beef patties has also been investigated. Maca and others (1997a) looked at different combinations of sodium lactate (0, 2, 3, or 4%), sodium propionate (0.1 or 0.2%), sodium acetate (0.1, 0.2, or 0.3%), and sodium citrate

(0.1, 0.2, or 0.3%) on the microbiological and sensory characteristics of vacuum packaged ground beef patties stored for 28 d at 4 °C. Treatments formulated with 3 or 4% sodium lactate with or without sodium propionate exhibited a bacteriostatic influence with similar sensory benefits as seen in top round roasts. Sodium acetate and sodium citrate treatments were ineffective at controlling the microbial population in this product. Eckert and others (1997) investigated the effect of sodium lactate (0, 3, or 4%) with or without sodium propionate (0.1 or 0.2%) on the microbial, sensory and chemical properties of aerobically stored hamburger patties held at 4°C for up to 3 days. Sodium lactate was bacteriostatic with and without the addition of propionate and propionate at 0.2% addition increased this effect. The combination of lactate and propionate at the highest levels tested decreased lipid oxidation slightly. Although sensory attributes did not change during storage, cooked hamburger formulations with lactate included had consistently higher beefy/brothy, beef fat, and sweetness scores, and those with both lactate and propionate had higher springiness and cohesiveness scores. Sodium propionate alone increased perceived juiciness scores.

An article by Shelef (1994) argues the potential antimicrobial mechanism of lactate. The mechanism is not well understood and needs additional investigation, particularly considering the increased antimicrobial action when used in combination with diacetate and other organic acids. It is likely that sodium and potassium lactate act similarly to other organic acids. The lactate molecule passes across the bacterial cell membrane in the undissociated (protonated) state and then dissociates (deprotonates) in the cell cytoplasm acidifying the cell interior. Growth becomes static because the

bacteria devotes much of the cell's available energy towards pumping protons out of the cell instead of supplying energy for cell growth and division. This mechanism was argued against by Young and Foegeding (1993) due to the fact that intracellular pH accounted for a portion but not all bacteriostatic activity. Although the addition of lactates to food products lowers water activity, is not believed to be a significant contributor to antimicrobial activity. The mechanism explaining the potent antimicrobial effects of lactate with diacetate against *L. monocytogenes* is also not understood and needs additional study. A number of possibilities exist including the potential of low diacetate concentrations altering cell membrane permeability, consequently making it difficult to regulate appropriate cytoplasmic pH. The addition of lactate/diacetate can also alter redox potential creating unfavorable conditions for listerial growth. It is also possible that high intracellular levels of lactate may interfere with critical metabolic precursors important for metabolism. Again, more research is needed to better understand this antimicrobial mechanism.

Irradiation of RTE Meats

Irradiation is a safe and effective antimicrobial treatment that kills viable pathogens such as *L. monocytogenes* in food products (Olson 1998). A limited amount of research is available on irradiated frankfurters. Terrell and others (1981b) investigated the sensory properties of irradiated (0, 0.8, and 3.2 kGy at either -34.4 or -51.5 °C) frankfurters made from pork (60%) and beef (40%), mechanically deboned chicken (100%), or mechanically deboned turkey (100%), all vacuum packaged. Co⁶⁰ irradiation was detrimental at all levels to overall sensory quality, flavor, texture, palatability and

color for all formulations. The severity was directly related to irradiation dose. Terrell (1981a, 1982) also demonstrated similar quality losses in frankfurters formulated with nitrite (50 ppm), nitrate (100 ppm), alpha tocopherol (206 ppm), pyrophosphate (3750 ppm) and then irradiated at the same dosage and temperatures as above. None of these treatments prevented a decline in quality attributes, and sensory palatability scores remained at unacceptable levels.

More recently, Sommers and Thayer (2000) determined that D_{10} -values for *L. monocytogenes* were dependent on the composition of the meat used in a frankfurter formulation. On the average, the D_{10} -value was 0.61 kGy for low irradiation levels. Foong and others (2004) reported the anti-listerial effect of irradiation on different processed meat products including frankfurters, ham, roast beef, bologna, and smoked turkey that were vacuum packaged and stored at refrigeration temperatures. D_{10} -values for products ranged from 0.55 to 0.58 kGy for *L. monocytogenes* and varied somewhat between product type. Initial reductions were not evident for irradiation treatments until after 4-6 wk storage. Quality attributes of irradiated (0 to 3.0 kGy) beef bologna formulated with various levels of added glucose (0, 2, 4, 6, or 8%) were determined by Sommers and Fan (2002). Added glucose did not change the anti-listerial effectiveness (D_{10} values of 0.59 to 0.60), but did change the quality attributes. Antioxidant activity increased with dextrose concentration, but no protective effect was seen in lipid oxidation values which increased with increasing irradiation dose from about 1.75 at 0 kGy to 3.5 at 4 kGy. Irradiation induced color changes, including loss of redness,

occurred with increasing irradiation dose. The a^* value declined from 14.4 at 0 kGy to 13.5 at 4 kGy.

The mechanism of cellular injury and death due to irradiation is well accepted as irradiation induced DNA damage (Olson 1998). This damage is generally a result of the generation of free radical radiolytic products of water molecules which readily react to break bonds within the DNA helical backbone structure which cannot readily repaired by cellular mechanisms and consequently inhibits transcription and protein synthesis. Secondary radiolytic products can also cause damage to a lesser extent.

Combined Intervention Strategies

The concept behind hurdle technology (combined interventions) is simple; the more fronts upon which a microorganism is attacked, the less chance it has to survive. Multiple interventions can have an additive or synergistic bactericidal/bacteriostatic effect while minimally affecting food quality. The combination of lactate/diacetate and pasteurizing doses of irradiation are effective at inhibiting the growth of *L. monocytogenes* on frankfurters (Sommers 2003 personal communication). Several studies have demonstrated the efficacy of antimicrobial additives in combination with irradiation against the outgrowth of *L. monocytogenes* on frankfurters.

Sommers and Fan (2003) and Sommers and others (2003a) investigated the use of diacetate (0.125 and 0.5 % of formulation) and citric acid (1, 5, and 10 % dips) in conjunction with irradiation (0.4 to 3.0 kGy). Both ingredients, in combination with irradiation, were effective for inhibiting the growth of *L. monocytogenes* initially and throughout storage. D_{10} -values of *L. monocytogenes* inoculated onto frankfurters

surfaces and then dipped into 0, 0.125, 0.25 and 0.5% sodium diacetate were 0.8, 0.53, 0.54 and 0.52 kGy, respectfully, while values for 0, 1, 5, or 10% citric acid were 0.61, 0.60, 0.54, and 0.53 kGy. Irradiation lethality was enhanced by the added ingredients with minimal changes in color, lipid oxidation, firmness, and antioxidant activity. However, no sensory evaluations were performed to determine sensory differences. Similar results were seen again by Sommers and others (2003b) who evaluated the relationship between potassium lactate and sodium diacetate incorporation into beef bologna and irradiation doses of 0, 1.5, or 3.0 kGy. Incorporation of the antimicrobials decreased the irradiation dose required for a one log reduction of *L. monocytogenes*. The D₁₀-value for frankfurters without incorporated antimicrobials was 0.56, while treatments with 1% lactate and 0.07% diacetate or 2% lactate and 0.15% diacetate had D₁₀-values of 0.53 and 0.46, respectively.

Regulatory Issues

Irradiation of RTE meat and poultry is not yet approved for control of microbiological hazards by the United States Department of Agriculture or the United States Food and Drug Administration, however, the need and value of irradiation is recognized by these organizations and future approval is expected. The body of scientific research on the safety and efficacy of irradiated RTE meat and poultry does not indicate potential barriers to approval or evidence of unsafe products, particularly when used in combination with other antimicrobial intervention techniques.

Currently, the interim rule for control of *Listeria monocytogenes* (9 CFR 430) in the Federal Register (USDA-FSIS 2004) outlines three alternative strategies for the

safety of RTE meat and poultry manufacturers. The first strategy states that a manufacture applies a post lethality treatment in addition to an antimicrobial agent or process to control the organism (such as lactate/diacetate *in combination with* irradiation). Option two involves the use of either a post-lethality treatment or an antimicrobial agent or process to control of *L. monocytogenes* (such as lactate/diacetate *or* irradiation *separately*). The third relies strictly on a processing facilities sanitation measures for control and does not require the application of any post-lethality treatment or additional antimicrobial hurdle (*neither* lactate/diacetate *nor* irradiation). Alternatives one and two rely on an antimicrobial or process to eliminate *L. monocytogenes* or limit it's growth if present. The organism may not increase to detectable levels during the products shelf-life or to levels that may result in a public health hazard. The rigor of protection against *L. monocytogenes* decreases from alternate one to alternate three.

Another regulatory issue that is impacted by this research is the zero tolerance rule for *L. monocytogenes*. There is significant debate on the stringency and usefulness of the rule based on a number of arguments including the fact that product treatments such as lactate/diacetate and irradiation can be used to manufacture RTE meat and poultry with a high degree of safety. More controversy exists over the policy itself with opponents arguing that it is unrealistic, impractical, and really doesn't decrease risk of listeriosis. In the future, the zero tolerance rule may need to be modified to account for the development of new antilisterial technologies.

CHAPTER III

MATERIALS AND METHODS

Treatments and Experimental Design

Phase 1

Frankfurters (~28% fat) formulated to contain a blend of beef (90/10 course ground lean, (Ruffino Meats and Food Service, Bryan, Texas, U.S.A.), pork (50/50 pork trimmings, (Ruffino Meats and Food Service), and mechanically deboned chicken (20% fat without added nitrite/nitrate or salt), (Tyson Foods, Inc., Springdale, Arkansas, U.S.A.) were produced with either no added lactate or a 3% lactate/diacetate solution (60% w/v commercially available to give final concentrations of 1.68% potassium lactate and 0.12% sodium diacetate in a completely processed frankfurter) (OptiForm PD 4, PURAC Biochem., The Netherlands) (Table 1). Meat raw materials were ordered for each processing day and randomly divided between both ingredient treatments. Processing of a standardized frankfurter formulation was performed at the Rosenthal Meat Science and Technology Center, a commercial-scale pilot plant located at Texas A&M University. The resulting product was vacuum packaged in pouches approved for irradiation (Cryovac B2570, Sealed Air Corp., Duncan, South Carolina, U.S.A) and arranged in 2 layers comprising a package of 8 frankfurters/pouch. After packaging, all samples for microbial, chemical, and sensory analysis were refrigerated, transported to the National Center for Electron Beam Food Research irradiation facility located at Texas A&M University's Research Park and irradiated to doses of 0, 1.8 or 2.6 kGy. Zero kGy dose samples (control) were passed through the irradiation chamber,

Table 1. Frankfurter formulation treatments prepared for inoculation with *Listeria monocytogenes* after irradiation and stored at 4 °C^a.

Ingredient ^c	Irradiation ^b		
	Control	1.8	2.6
Control	X	X	X
3% Lactate/Diacetate	X	X	X

^aStored at 4 °C with sampling intervals of 0, 1, 2, 3, 4 wk.

^bLevels of irradiation were derived from Sommers (2003) Sommers and others (2003b), 1.8 kGy will result in a 3 log cfu reduction and 2.6 kGy will result in a 5 log cfu reduction.

^cFrankfurters were formulated with 0% (control) or 3% lactate/diacetate solution.

but no irradiation treatment was applied. Frankfurter packages were irradiated inside a single layer cardboard box. Dual beam irradiation was used to minimize dose variation throughout the sample. High density polyethylene sheets were used as attenuators for reducing the energy of incident electrons to achieve the target doses. Dose mapping of frankfurter packages was performed using alanine pellets (Harwell Dosimeters, U.K.) placed in the top, bottom, and middle of packages, which were then irradiated and analyzed for absorbed dose using electron paramagnetic resonance spectroscopy (Bruker EMS 104 EPR Analyzer, Bruker Instruments, Germany). Multiple dosimetry runs were performed to finalize the exact attenuation scheme and conveyor speed required for the desired electron beam doses of the experiment. After irradiation, samples destined for pathogen inoculation were transferred immediately to the Food Microbiology Laboratory located in the Kleberg Center at Texas A&M University. Packages for all treatments, including non-inoculated frankfurters, were opened using sterile technique. The surface of each frankfurter destined for microbial analysis was inoculated with 0.1 ml of a cocktail of rifampicin resistant *Listeria monocytogenes* strains 15313, 51414, 43256, and 74166 (American Type Culture Collection, Manassas, Virginia, U.S.A). The frankfurter surface then was allowed to dry for 1 h and aerobically packaged.

Strains of rifampicin resistant *L. monocytogenes* were prepared according to procedures described by Kaspar and Tamplin (1993) and maintained on tryptic soy agar (Difco Laboratories, Detroit, Michigan, U.S.A.) slants. Inoculum was prepared by aseptically adding a loop of culture to separate tryptic soy broth tubes (Difco

Laboratories) and incubating overnight. Log phase culture strains were combined and serially diluted in 0.1% bacto-peptone to appropriate concentrations to achieve the desired final inoculation level. The inoculum was added to yield approximately 10^3 cfu/frankfurter. This inoculation level was based on the maximum incidence levels of *L. monocytogenes* on commercial frankfurters reported by Wallace and others (2003) and enabled counts indicative of growth or a decline in pathogen numbers. Samples destined for chemical/physical and sensory analyses were not inoculated. All samples were stored at 4 °C and analyzed after 0, 1, 2, 3, and 4 wk of aerobic storage (representative of an open package in a consumer's refrigerator). Comparisons of treatments were made with the non-irradiated control at the designated sampling intervals.

Phase 2

Frankfurters (~28% fat) were formulated and manufactured as described in Phase 1 and transferred to the Food Microbiology Laboratory located in the Kleberg Center adjacent to the Rosenthal Center on the Texas A&M University campus. Packages destined for pathogen inoculation were surface inoculated with 0.1 ml of a cocktail of rifampicin resistant *Listeria monocytogenes* strains 15313, 51414, 43256, and 74166 (American Type Culture Collection) to yield approximately 10^5 cfu/frankfurter (Table 2). The surface was allowed to dry for 1 h, the packages vacuum sealed in a barrier bag approved for irradiation (Cryovac B2570), and sealed in another barrier bag to meet the electron beam facility requirements for samples inoculated with known pathogens. Samples were transported to the National Center for Electron Beam Food Research and

Table 2. Frankfurter formulation treatments prepared for inoculation with *Listeria monocytogenes* prior to irradiation and stored at 4 °C^a.

Ingredient ^c	Irradiation ^b		
	Control	1.8	2.6
Control	X	X	X
3% Lactate/Diacetate	X	X	X

^aStored at 4 °C with sampling intervals of 0, 2, 4, 6, 8 wk.

^bLevels of irradiation were derived from Sommers (2003) and Sommers and others (2003b), 1.8 kGy will result in a 3 log cfu reduction and 2.6 kGy will result in a 5 log cfu reduction.

^cFrankfurters were formulated with 0% (control) or 3% lactate/diacetate solution.

irradiated to doses of 0, 1.8 or 2.6 kGy as in Phase 1. Samples destined for chemical and sensory analysis were not inoculated. All samples were then held at 4 °C and analyzed after 0, 2, 4, 6 and 8 wk of storage. Comparisons of treatments were made with the non-irradiated control at the designated sampling intervals.

Methods of Analysis

Microbial Analyses

Aerobic Plate Counts (APC) were determined using the enumeration procedures described by Messer and others (1985). On each sampling day, one frankfurter pouch was sampled and one frank removed and placed in a Stomacher bag (Seward, Inc., U.K.) with 25ml of 0.1% Bacto[®] Buffered Peptone Water (BPW) (Difco Laboratories). The contents of each bag were vigorously massaged by hand for 2 min without disrupting the frankfurter structure. Microbial counts as log cfu/frankfurter were enumerated from 10 fold dilutions of this homogenate in 9 ml test tubes of 0.1 % BPW and plated onto Petrifilm[™] Aerobic Count plates (3M Health Care Microbiology Products, St. Paul, Minnesota, U.S.A.). The plates were incubated at 30 °C for 48 h and counted according to the Petrifilm interpretation guidelines using a Quebec Darkfield Colony Counter (Leica Inc., Buffalo, New York, U.S.A.).

Listeria monocytogenes was enumerated according to Ryser and Donnelly (2001) by direct plating onto Rif-TSA (Rifampicin- Tryptic Soy Agar [100 µg rifampicin per ml media] Sigma Chemical Company, St. Louis, Missouri, U.S.A. and Difco Laboratories) using the same sampling and dilution procedure described for APC. Data was presented as log *L. monocytogenes* cfu/frankfurter.

Chemical/Physical Analyses

To estimate degree of lipid oxidation, 2-thiobarbituric acid reactive substances (TBARS) were determined using the procedure for cured meats as described by Tarladgis and others (1960) and modified by Rhee and Ziprin (1981). Sixty g of sample was blended with 87 ml dd water, 3 ml of sulfanilamide solution (0.5% in 20% hydrochloric acid v/v, Sigma Chemical Company), and 30 ml of propyl gallate (0.5% w/v, Sigma Chemical Company) with ethylenediaminetetraacetic acid (0.5% w/v, Sigma Chemical Company) solution in water. One blending was performed per 60 g sample. Thirty g of this homogenate was added to 77.5 ml dd water and 2.0 ml of 4 N hydrochloric acid (EM Industries Inc., Hawthorne, New York, U.S.A.). This was distilled to a volume of 50 ml and two distillations were performed per blending. For development of color, 5 ml of distillate was combined with 5 ml of TBARS solution (0.02 M TBARS in ddwater, Sigma Chemical Co.), heated to 100°C for 35 min, and cooled to ambient temperature. Duplicate color developments were made for each distillation, and absorbance of the resulting solution was measured at 530 nm using a Cary 300 Dual Beam Spectrophotometer (Varian Analytical Instruments, Palo Alto, California, U.S.A.). Five ml of the TBARS solution heated with 5 ml ddwater served as a blank. Reported TBARS values were calculated as mg of malonaldehyde per kg of sample and expressed as a TBARS, where $\text{TBARS} = \text{Absorbance @530 nm} \times 7.8$ (conversion factor).

pH was determined by a slurry method adapted for meat products using an Orion™ (model 720A, Orion Research, Inc., Beverly, Massachusetts., U.S.A.) pH meter

(pre-calibrated using pH 4 and 7 standards) and combination electrode. Thirty g of homogenized sample was blended with 90 ml of DI water for 2 min in a Waring® Blender (Model 31BL92, Waring Products Division, Dynamic Corp. of America, New Hartford, Connecticut., U.S.A.) and the pH probe inserted into the stirred slurry for measurement.

Proximate analysis included percentage of moisture, lipid and total protein as determined according to AOAC (2000) procedures. Moisture content was assessed using the CEM Smart Trac System 5 (microwave determination) and lipid content using the CEM AES-9001 Extraction System (dichloromethane extraction) (CEM Corporation, Matthews, North Carolina., U.S.A.). Total nitrogen was analyzed using the Leco FP528 Nitrogen Determinator (Leco Corp., St. Joseph, Michigan, U.S.A.), standardized against an oatmeal standard and the total protein determined by multiplying the percent nitrogen by a 6.25 conversion factor.

$L^*a^*b^*$ color space values for the interior and exterior surfaces of each product were obtained by reflectance using a Minolta Colorimeter (CR-200, Minolta Corp., Ramsey, New Jersey, U.S.A.). The colorimeter was standardized before each use using a white Minolta calibration plate (C values: $Y=92.5$, $x=0.3136$, and $y=0.3196$). L^* color space values indicated the degree of whiteness (100) to blackness (0). Positive a^* values indicated the degree of redness whereas negative a^* values indicated greenness. Positive b^* values indicated yellowness while negative b^* values indicated blueness. For each measurement, the colorimeter port was covered with clear Saran Wrap® and random

readings were taken at 6 locations on the inner and outer surfaces of the frankfurter after appropriate calibration of the instrument. Color space values were reported as $L^*a^*b^*$.

Sensory Analyses

Descriptive attribute analysis was performed on uninoculated products and evaluated by an expert flavor and texture descriptive attribute sensory panel at the Texas A&M University Sensory Testing Facility. Panelists were selected and trained according to the procedures of AMSA (1995); Cross and others (1978); and Meilgaard and others (1991). Training prior to testing was conducted by presenting reference samples to the panel to characterize the basic attributes of the various products (3 training sessions). A sample sensory ballot that listed all attributes evaluated is included in Appendix B. Samples were evaluated for aroma and flavor, basic tastes, feeling factors, mouth feel, aftertastes, and texture attributes using the 16 point Spectrum™ Universal Intensity Scale (Meilgaard and others 1991) where 0 = absence of an attribute and 15 = extremely intense. Individual sensory attributes are described in Appendix C.

Samples were cooked by steeping in boiled distilled water for 7 min and immediately cut into 1.5 cm pieces. Three sections of frankfurter of each treatment were served to the six member trained sensory panel. Panelists evaluated samples in random order and samples were labeled with random three digit numbers. Twelve treatments and one warm-up sample were evaluated over a 2 h period with one, 15 min break between sessions. Panelists evaluated the samples for all attributes in isolated booths fitted with a breadbox server and red incandescent lighting to mask color differences. Unsalted crackers, Ricotta cheese, and distilled water were available to panelists to cleanse the

pallet between samples. Interior and exterior color evaluations were conducted under florescent white light using a color swatch standard scale developed by the panelists (Tables 3 and 4).

Statistical Analyses

A total of three replications per phase were performed for statistical validity and the statistical analysis was arranged as a complete block design. Analysis of variance was performed using the General Linear Model (GLM) procedures of the Statistical Analysis System (SAS 1995). Analysis of variance determined statistical differences among the main effects and interactions at a significance level of $p < 0.05$. Least squares means were calculated to identify significant treatment effects using the pdiff function of SAS.

The frankfurters produced for these study were an extremely homogeneous and highly emulsified product with very little background variation. On each processing day, meat for all treatments was ground in one bulk batch and then randomly divided between all treatments. For sensory analyses, panelist interactions were evident for nearly every significant attribute. To this end, statistical exercises were performed to ensure the validity of the highly trained panel for sensory data sets. In light of this variation, significant panelist interactions were graphed to evaluate and determine their cause. In all cases, small shifts in individual panelist scores over time or between different panelists were the reason for significant panelist interactions. It was decided to condense the model statement to remove panelist effect. This allowed for a much clearer indication of significant differences between other treatment variables.

Table 3. L*a*b* and descriptions of color swatch references used by panelists to evaluate frankfurters.

Location	Score	L*	a*	b*	Panelist description of the color
Exterior	8	40.66	15.33	20.34	Mahogany
	7	42.65	19.06	23.18	Chocolate orange
	6	44.08	22.75	31.65	Rusty red
	5	50.38	21.80	31.83	Burnt orange
	4	54.39	22.40	35.10	Terra cotta orange
	3	57.40	19.27	36.64	Matte peachy bronze
	2	64.75	16.51	34.67	Peachy tan
	1	69.01	14.47	33.79	Fleshy tan
Interior	8	48.47	24.75	27.23	Rust
	7	54.31	25.42	28.96	Pale rust
	6	58.88	23.74	26.65	Light orange brown
	5	68.64	20.98	24.57	Apricot brown
	4	78.04	13.41	18.27	Apricot
	3	77.05	16.53	14.76	Pale rose
	2	83.05	9.65	10.59	Fleshy pink
	1	83.81	9.26	11.94	Light pink

Table 4. Color swatch references used by panelists to evaluate frankfurters.

Location	Score	Brand	Manufacturers Code	Name
Exterior	8	Behr Premium Plus	230F-7	Florence Brown
	7	Valspar	298-6	Western Sky
	6	American Tradition	2003-7B	La Fonda Deep Clay Red
	5	Valspar	266A-6	California Copper
	4	Valspar	266A-5	Copper Hill
	3	Valspar	266A-4	Ginger Root
	2	Valspar	266A-3	Kenyan Tan
	1	Valspar	266A-2	Fairhaven Peach
Interior	8	Valspar	298-6	Western Sky
	7	American Tradition	2003-7B	La Fonda Deep Clay Red
	6	Valspar	298-6	Stallion
	5	Valspar	298-4	Buckskin
	4	Valspar	298-2	Formosa
	3	American Tradition	2005-8B	Resort Peach
	2	American Tradition	2005-8C	Butterfly Pink
	1	Olympic Paints	B21-1	Sand Castle

CHAPTER IV

MICROBIAL RESULTS AND DISCUSSION

Phase 1

Phase 1 monitored the growth of *L. monocytogenes* inoculated onto the surface of frankfurters before application of irradiation treatment followed by aerobic storage. This simulated contamination by a consumer after storage and subsequent aerobic growth conditions in an opened package in the consumer's refrigerator. Overall, the incorporation of lactate/diacetate into frankfurter formulations had a strong bacteriostatic effect on microbial counts throughout storage under aerobic conditions. Irradiation had a small, but significant influence on microbial counts at later storage dates.

Figure 1 summarizes aerobic plate counts (APC) for phase 1. Initial APC values were not significantly different between treatments with an average value of 5.18 cfu/frankfurter. Aerobic Plate Counts increased steadily throughout storage for frankfurters formulated without lactate/diacetate to over 9.0 cfu/frankfurter regardless of irradiation treatment. However, APC counts of frankfurters formulated with lactate/diacetate remained similar to initial counts with a value of 6.5 cfu/frankfurter for the non-irradiated control and 4.73 and 4.97 cfu/frankfurter for 1.8 and 2.6 kGy irradiated frankfurters respectively. Specifically, microbial counts did not differ between treatments until storage wk 2 when APC counts for non-irradiated frankfurters formulated without lactate/diacetate reached significantly higher counts than all other treatments. All lactate/diacetate formulated frankfurters had significantly lower APC counts than those

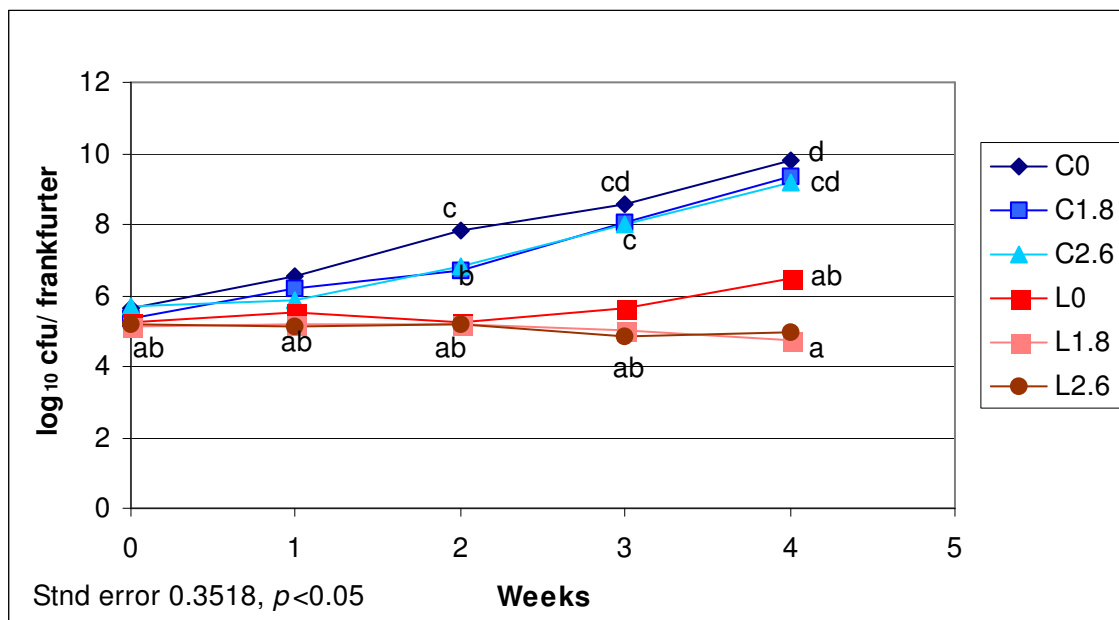


Fig. 1. APC counts (log cfu/frank) of frankfurters formulated with or without lactate/diacetate, treated with electron beam irradiation before inoculation, and stored aerobically at 4 °C. Means with different subscripts are significantly different ($p < 0.05$). (Legend terms: C= no lactate/diacetate in formulation, L= 3% lactate/diacetate solution addition, 0= no irradiation treatment, 1.8 and 2.6= applied irradiation dose in kGy).

without lactate/diacetate at wk 3 regardless of irradiation treatment and that trend continued throughout storage. There was a significant irradiation effect at wk 3 and 4 when non-irradiated frankfurters formulated with lactate/diacetate had a significantly higher APC values than both irradiated frankfurters formulated with lactate/diacetate.

Figure 2 summarizes *L. monocytogenes* counts for phase 1. Initial counts of *L. monocytogenes* were not significantly different between treatments with an average log value of 5.30 *L. monocytogenes* cfu/frankfurter. Significantly different counts were noted on wk 2 and continued through wk 4 of storage when all lactate/diacetate formulated frankfurters had significantly lower counts (5.45 cfu average) than those without lactate/diacetate (9.07 cfu average). Irradiation alone had little influence on suppressing *L. monocytogenes* growth, which increased from 5.4 to 9.3 log cfu in both the irradiated and control frankfurters.

The addition of lactate/diacetate to a frankfurter formulation was an effective method of controlling the outgrowth of *L. monocytogenes* aerobically while irradiation had limited antimicrobial influence during storage. For both APC and *L. monocytogenes*, no initial irradiation effects were seen since irradiation occurred before inoculation. These microbial intervention methods have important food safety implications. If a consumer accidentally contaminates a package of frankfurters with *L. monocytogenes*, there is little protection and the organism can grow to high numbers. The aerobic growth of *L. monocytogenes* on RTE meats has not been well studied despite the fact that it plays an important role in the microbial safety of these products. This is presumably due to the fact that nearly all frankfurters manufactured in the United States have a plastic

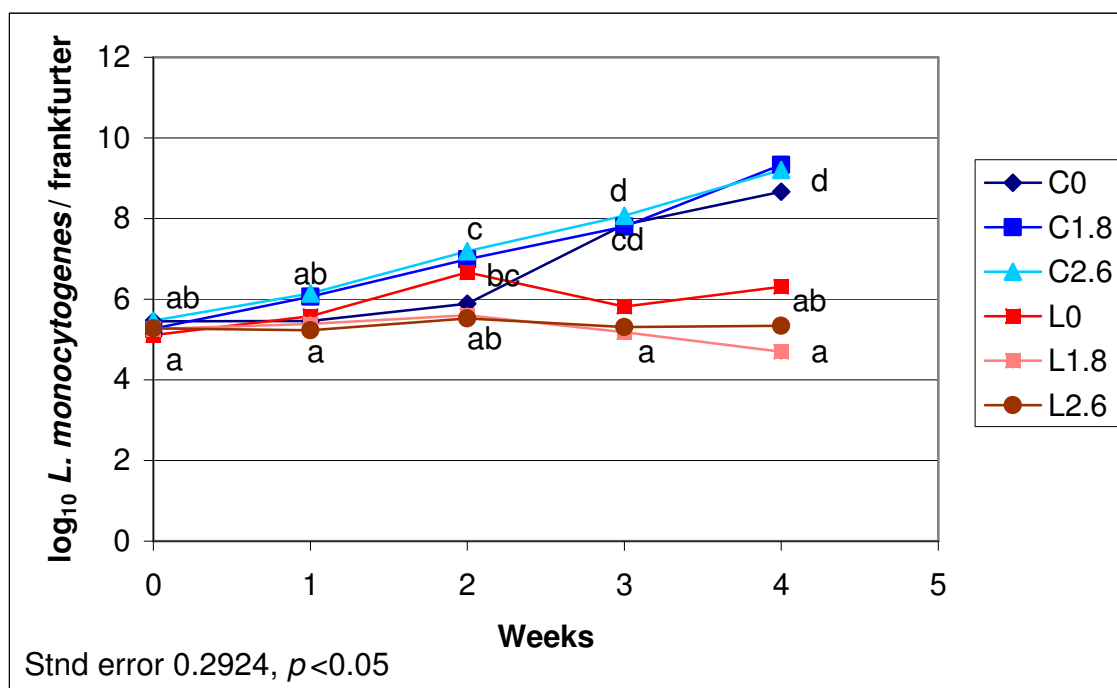


Fig. 2. *L. monocytogenes* counts ($\log L. monocytogenes/\text{frank}$) of frankfurters formulated with or without lactate/diacetate, treated with electron beam irradiation before inoculation, and stored aerobically at 4 °C. Means with different subscripts are significantly different ($p < 0.05$). (Legend terms: C= no lactate/diacetate in formulation, L= 3% lactate/diacetate solution addition, 0= no irradiation treatment, 1.8 and 2.6= applied irradiation dose in kGy).

vacuum sealed pouch format. Even so, the typical consumer opens the sealed vacuum packaged pouch and in doing so, creates an aerobic environment that the frankfurters may be subjected to until spoilage occurs.

Phase 2

Phase 2 monitored the growth of *L. monocytogenes* inoculated onto the surface of frankfurters, followed by vacuum packaging, the application of irradiation and storage at 4 °C. This simulates contamination after the processing cooking step and subsequent growth in a sealed package before consumer use. Overall, both irradiation and the addition of lactate/diacetate to a frankfurter formulation were effective towards controlling microbial growth of *L. monocytogenes* in an unopened frankfurter package.

Figure 3 summarizes APC values for phase 2. Similar values were seen initially for each irradiation treatment with an average value of 7.57 cfu/frankfurter for frankfurters treated with 0 kGy. Irradiation treatments reduced initial total microbial loads with the application of 1.8 kGy resulting in a 3.21 cfu/frankfurter average reduction and the application of 2.6 kGy resulting in a 5.2 cfu/frankfurter average reduction. Throughout storage, lactate/diacetate in combination with irradiation had a bacteriostatic effect, however, APC values for irradiated frankfurters without lactate/diacetate gradually increased during storage even though irradiation treatments initially suppressed growth. Specifically, APC values for frankfurters formulated without lactate/diacetate and irradiated to 1.8 kGy began to have significantly higher counts at 6 wk and those irradiated to 2.6 kGy at 8 wk. Frankfurters that were formulated with lactate/diacetate and irradiated with 2.6 kGy had the lowest APC values of all treatments

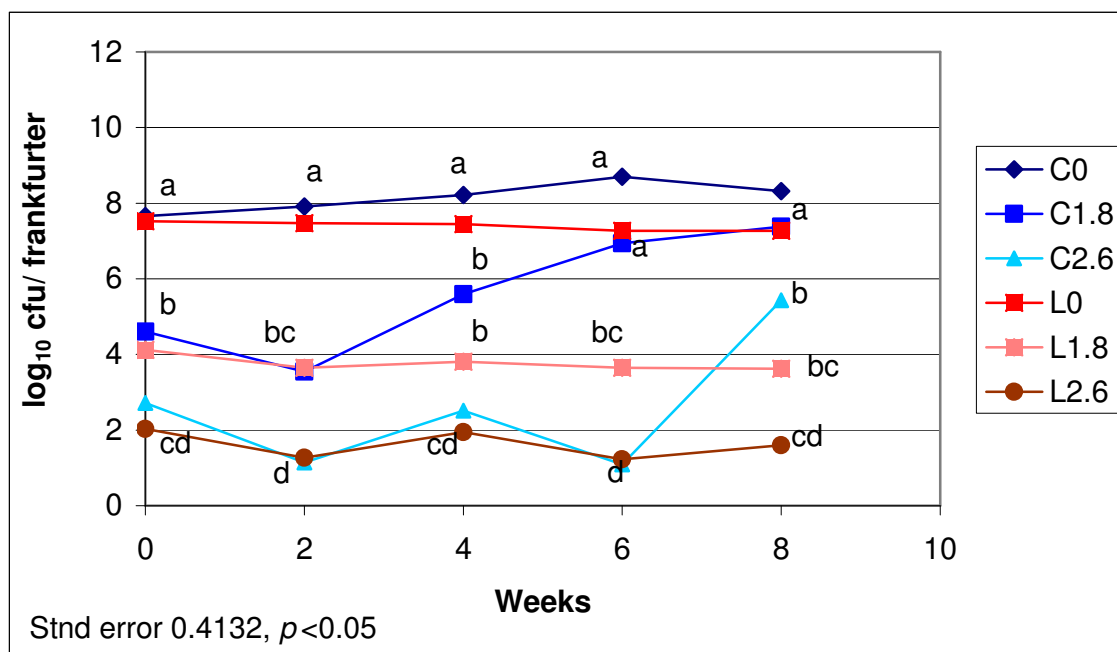


Fig. 3. APC counts (log cfu/frank) of frankfurters formulated with or without lactate/diacetate, treated with electron beam irradiation after inoculation, and stored vacuum packaged at 4 °C. Means with different subscripts are significantly different ($p < 0.05$). (Legend terms: C= no lactate/diacetate in formulation, L= 3% lactate/diacetate solution addition, 0= no irradiation treatment, 1.8 and 2.6= applied irradiation dose in kGy).

while those receiving 1.8 kGy tended to be intermediate throughout the 8 wk storage period.

Figure 4 summarizes *L. monocytogenes* counts for phase 2. As seen with the APC values, initial *L. monocytogenes* counts at 0 kGy averaged 7.63 cfu/frankfurter. Irradiation treatments significantly reduced initial *Listeria* counts. With the application of 1.8 kGy, log counts were reduced by an average of 3.17 cfu/frankfurter while the application of 2.6 kGy resulted in a 5.01 cfu/frankfurter average reduction. As seen with APC values, there was a listeristatic effect that was maintained throughout storage for all irradiated frankfurters formulated with lactate/diacetate, while those without lactate/diacetate tended to have increased growth after 8 wk. Specifically, *L. monocytogenes* values for frankfurters formulated without lactate/diacetate and irradiated to 1.8 kGy had significantly higher counts after 4 wk storage. Frankfurters formulated without lactate and irradiated to 2.6 kGy likewise had higher *L. monocytogenes* counts at wk 8 of storage when compared to wk 6. Frankfurters that were formulated with lactate/diacetate and irradiated with 2.6 kGy had the lowest *L. monocytogenes* values at 8 wk while counts for 1.8 kGy were intermediate.

These results were similar to a number of studies that demonstrated the listeristatic efficacy of lactate and/or diacetate incorporated into the formulation of comminuted products. As observed by Barmpalia (2004); Mbandi and Shelef (2001,2002); Nunez (2004a); Samelis and others (2002); and Sommers and others (2003b), 3% sodium or potassium lactate particularly in combination with sodium diacetate, have a listeristatic effect in a variety of processed meat products throughout

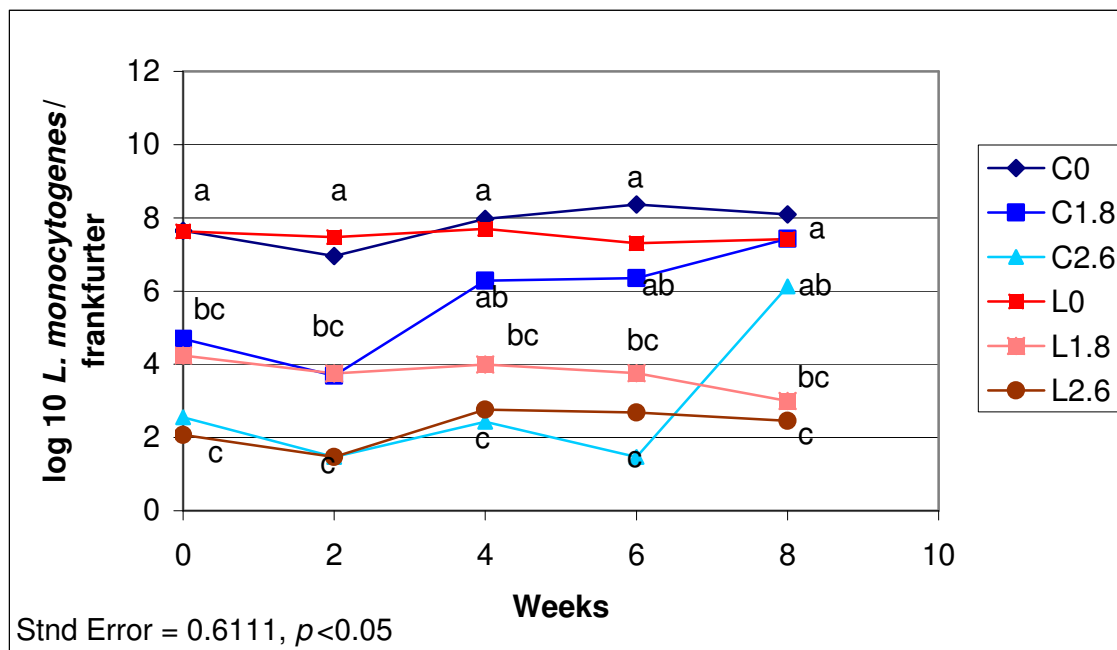


Fig. 4. *L. monocytogenes* counts (log *L. monocytogenes*/ frank) of frankfurters formulated with or without lactate/diacetate, treated with electron beam irradiation after inoculation, and stored vacuum packaged at 4 °C. Means with different subscripts are significantly different ($p < 0.05$). (Legend terms: C= no lactate/diacetate in formulation, L= 3% lactate/diacetate solution addition, 0= no irradiation treatment, 1.8 and 2.6= applied irradiation dose in kGy).

refrigerated storage. In these studies, some shelf-life tests lasted for up to 120 d. Table 5 shows the calculated initial D_{10} values of *L. monocytogenes* when treated with electron beam irradiation. D_{10} -values for control frankfurters ranged from 0.56 to 0.57, while frankfurters formulated with lactate/diacetate had a D_{10} -value of 0.51. While the initial log cfu values are not significantly different, the D_{10} -values indicate that incorporation of lactate/diacetate into a frankfurter formulation may slightly decrease the irradiation dose required to kill 90% of *L. monocytogenes* on frankfurters when measured as APC or with selective Rif-TSA media. These D_{10} -values are comparable to those of Sommers and others (2003b) and Sommers and Thayer (2000) who observed a decrease in D_{10} -value from about 0.59 to 0.53-0.46 for treatments with comparable amounts of lactate/diacetate incorporated into frankfurter formulations.

In general, lactate/diacetate incorporation was effective for preventing outgrowth of *L. monocytogenes* in an unopened frankfurter package. Pasteurizing doses of irradiation were also effective for reducing the microbial load on frankfurters. The inoculation level used for frankfurters in this study was high compared to contamination levels reported on commercially available product. Theoretically, pasteurizing doses of irradiation should reduce the level of contamination to virtually zero. If small numbers of the organism do survive, the lactate/diacetate treatment would prevent outgrowth. Therefore, the use of these two treatments in combination should be effective for ensuring the microbial safety of frankfurters.

Table 5. D₁₀-values (kGy) for *L. monocytogenes* inoculated onto the surface of frankfurters formulated with or without lactate/diacetate and irradiated.

Ingredient ^a	APC	<i>L. monocytogenes</i>
Control	0.56	0.57
Lactate	0.51	0.51
R ²	0.94	0.95

^aFrankfurters were formulated with 0% (control) or 3% lactate/diacetate solution and irradiated to 0, 1.8, or 2.6 kGy.

CHAPTER V

SENSORY RESULTS AND DISCUSSION

Phase 1

During phase 1, the sensory properties of irradiated frankfurters formulated with or without lactate/diacetate were monitored throughout aerobic storage.

Sensory Aroma

During phase 1, a highly trained sensory panel monitored changes in frankfurter aroma during aerobic storage. Panelists identified significant aromatics including meaty/brothy complex, fat complex, smoke, and spice complex. These were expected since major groups of volatile odor compounds in frankfurters include a variety of terpenes and phenolic compounds from spices and smoke (Chevance and Farmer 1999). Panelists looked for, but were not able to identify, beefy/brothy, porky/brothy, chicken/brothy, beef fat, pork fat, chicken fat, cardboard, painty, fishy, livery, caramelized, soured, soapy, and wet dog aromatics in significant amounts. Overall, the addition of lactate/diacetate had a protective effect for frankfurter aroma throughout storage, particularly near the end of storage when the aroma of frankfurters formulated without lactate/diacetate deteriorated. Irradiation had a small but significant effect on protecting quality during the product shelf-life and did not negatively impact frankfurter quality.

Meaty/brothy complex, smoke, and spice aroma attributes were judged by panelists to be significantly lower for frankfurters formulated without lactate/diacetate than for those with lactate/diacetate during the last storage week (Table 6). At 4 wk of

storage frankfurters without lactate/diacetate were judged to have less meaty/brothy (2.17), less smoke (0.51), and less spice complex aroma (1.26) than those formulated with lactate (2.74, 0.76, and 1.72 respectively). Differences between treatments were not identified at other times during storage. The decline in these attributes near the end of storage is consistent with product quality loss and the end of useable shelf-life.

Table 7 shows small, but significantly detectable differences, in smoke aroma between irradiation treatments for each storage week. Sensory smoke aroma scores were initially equal for all irradiation treatments (0.79 to 0.76), however the scores varied somewhat during the following week. There was an indication that irradiation might help protect loss of smoke aroma. The non-irradiated control at wk 4 had a lower (0.58) smoke score than both comparable irradiated frankfurters after 4 wk. However, at these low levels of aroma, it was difficult to distinguish differences.

Replication effects were evident for sensory aroma (Table 8). In the replication by week interactions, fat complex scores were higher in replication 1 and decreased as storage progressed. Replication 2 and 3 scores were initially low (0.4) and remained consistently low throughout storage. This may be related to unusually high TBARS values seen in replication 1, but not in replications 2 and 3. Table 8 also shows replication 1 to have a higher smoke aroma score than replications 2 and 3 for all storage weeks except wk 2, however, there was a great deal of inconsistency in the differences with no clearly defined trends. These replication differences may be due to smokehouse operation inconsistencies for different processing runs during which the natural smoke

Table 6. Least squares means of meaty/brothy, smoke, and spice complex aroma for irradiated frankfurters formulated with or without lactate and aerobically stored at 4 °C.^a

Wk	Meaty/brothy		Smoke		Spice complex	
	Control ^b	Lactate ^b	Control ^b	Lactate ^b	Control ^b	Lactate ^b
0	2.82 a	2.91 a	0.74 a	0.79 a	1.74 ab	1.89 a
1	3.04 a	3.02 a	0.91 a	0.88 a	1.86 ab	1.84 ab
2	2.69 a	2.63 a	0.74 a	0.76 a	1.78 ab	1.83 ab
3	2.64 b	2.77 a	0.73 a	0.81 a	1.49 b	1.64 ab
4	2.17 b	2.74 a	0.51 b	0.76 a	1.26 c	1.72 ab
Std error	0.0841		0.0444		0.0782	

^aMeans within a sensory parameter category with different letters are significantly different ($p < 0.05$).

^bFrankfurters were formulated with 0% (control) or 3% lactate/diacetate solution.

Table 7. Least squares means for smoke aroma for irradiated frankfurters formulated with or without lactate and aerobically stored at 4 °C.^a

Wk	Irradiation (kGy) ^b		
	Control	1.8	2.6
0	0.79 ab	0.76 ab	0.76 ab
1	0.98 a	0.87 a	0.83 ab
2	0.87 a	0.67 ab	0.70 ab
3	0.68 ab	0.87 a	0.77 ab
4	0.58 b	0.65 ab	0.67 ab
Std error	= 0.0544		

^aMeans with different letters are significantly different ($p < 0.05$).

^bNot irradiated (control) or irradiated to 1.8 or 2.6 kGy.

Table 8. Least squares means of fat complex and smoke aroma for irradiated frankfurters formulated with or without lactate and aerobically stored at 4 °C.^a

Wk	Fat complex			Smoke		
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
0	1.13 a	0.15 c	0.20 c	0.97 b	0.62 d	0.72 cd
1	1.15 a	0.17 c	0.21 c	1.22 a	0.57 d	0.90 bc
2	0.68 b	0.13 c	0.15 c	0.86 bc	0.71 cd	0.72 cd
3	0.78 b	0.17 c	0.13 c	1.03 b	0.55 d	0.73 cd
4	0.33 c	0.13 c	0.17 c	0.90 bc	0.48 d	0.52 d
Std error	0.0512			0.0544		

^aMeans within a sensory parameter category with different letters are significantly different ($p < 0.05$).

generators may not have been functioning correctly and therefore smoke application may have been inconsistent.

Overall, the addition of lactate/diacetate to a frankfurter formulation had a positive impact on frankfurter aroma as judged by a trained sensory panel. It was effective at maintaining product quality during late storage dates when frankfurters formulated without lactate began to deteriorate.

Sensory Flavor

Panelists identified several significant sensory attributes including aromatics (meaty/brothy complex, fat complex, smoke, and spice complex), feeling factors (metallic, astringent, and mouthburn), tastes (salt, sour, bitter, and sweet), and aftertastes (fat mouthfeel and smoke/spice complex). These attributes were expected to be present in frankfurters (Chevance and Farmer 1999). The panelists looked for, but were not able to identify, the aromatics beefy/brothy, porky/brothy, chicken/brothy, beef fat, pork fat, chicken fat, cardboard, painty, fishy, livery, caramelized, soured, soapy, and wet dog in significant amounts.

Overall, most flavor attributes were not strongly influenced by lactate/diacetate or irradiation treatments until the latter part of the storage period. Both lactate/diacetate and irradiation had varying effects on product flavor attributes. The protective effect of lactate/diacetate and irradiation treatments was evident for aromatics such as meaty/brothy complex, smoke/spice complex, and spice complex, and also for flavors like astringency, sourness, and bitterness. As shown in Tables 9 and 10, sensory scores for lactate/diacetate frankfurters remained consistent throughout storage, however, a

Table 9. Least squares means of meaty/brothy and smoke/spice complex aftertaste for irradiated frankfurters formulated with or without lactate/diacetate and aerobically stored at 4 °C.^a

Wk	Meaty/brothy		Smoke/spice complex	
	Control ^b	Lactate ^b	Control ^b	Lactate ^b
0	4.57 a	4.61 a	2.49 a	2.69 a
1	5.02 a	5.02 a	2.59 a	2.76 a
2	4.77 a	4.71 a	2.45 a	2.60 a
3	5.09 a	5.13 a	2.34 a	2.56 a
4	4.11 b	4.98 a	1.93 b	2.61 a
Std error	0.1354		0.0779	

^aMeans within a sensory parameter category with different letters are significantly different ($p < 0.05$).

^bFrankfurters were formulated with 0% (control) or 3% lactate/diacetate solution.

Table 10. Least squares means of spice complex flavor aromatics for irradiated frankfurters formulated with or without lactate/diacetate and aerobically stored at 4 °C.^a

Ingredient ^b	Irradiation ^c	Wk				
		0	1	2	3	4
Control	Control	2.86 a	2.92 a	2.68 a	2.67 a	1.40 c
	1.8	2.86 a	2.87 a	2.81 a	2.70 a	2.33 b
	2.6	2.64 a	2.86 a	2.63 a	2.70 a	2.37 b
Lactate	Control	2.96 a	3.16 a	3.11 a	3.20 a	3.00 a
	1.8	3.17 a	2.93 a	2.86 a	3.03 a	2.83 a
	2.6	3.23 a	3.04 a	2.87 a	2.93 a	2.80 a
Std error = 0.1363						

^aMeans with different letters are significantly different ($p < 0.05$).

^bFrankfurters were formulated with 0% (control) or 3% lactate/diacetate solution.

^cNot irradiated (control) or irradiated to 1.8 or 2.6 kGy.

decline in each attribute occurred at wk 4 for non-irradiated frankfurters formulated without lactate and compared to every other treatment. Specifically, meaty/brothy complex flavor scores declined to 4.11 at wk 4 of storage for frankfurters without lactate/diacetate compared to the non-irradiated frankfurter formulated with lactate which had a score of 4.98 (Table 9). The smoke/spice complex aftertaste score declined significantly at 4 wk of storage, while frankfurters formulated with lactate/diacetate had a score of 2.61 compared to a score of 1.93 for those without lactate/diacetate (Table 9). At wk 4 of storage, the spice complex aromatic score for non-irradiated frankfurters formulated without lactate/diacetate was 1.4 compared to a score of 3.0 for non-irradiated frankfurters with lactate/diacetate. This demonstrates a protective effect for the lactate/diacetate treatment (Table 10). A small decline was also noted on wk 4 for spice complex in irradiated frankfurters without lactate/diacetate. This deterioration in flavor toward the end of storage is likely indicative of quality decline usually associated with aerobic spoilage. A similar effect was seen for astringency, sourness, and bitterness (Tables 11 to 13). In general, although not always significantly different, lactate/diacetate formulated frankfurters were slightly more astringent, sour, and bitter than those without lactate/diacetate.

Overall, frankfurters formulated with lactate/diacetate were slightly more metallic in flavor and saltier (Table 14). Metallic flavor decreased slightly over storage for all treatments from 2.29 to 2.13 after 4 wk of storage (Table 15). The mixture of potassium lactate and sodium diacetate used in this study is described as having a mildly saline taste by the manufacturer and likely accounts for saltiness effect. Potassium

Table 11. Least squares means of astringency for irradiated frankfurters formulated with or without lactate/diacetate and aerobically stored at 4 °C.^a

Ingredient ^b	Irradiation ^c	Wk				
		0	1	2	3	4
Control	Control	2.62 ab	2.61 ab	2.43 b	2.47 b	2.03 c
	1.8	2.49 b	2.53 ab	2.27 b	2.33 b	2.47 b
	2.6	2.39 b	2.61 ab	2.48 b	2.27 b	2.47 b
Lactate	Control	2.77 a	2.74 a	2.78 a	2.57 ab	2.73 a
	1.8	2.77 a	2.73 a	2.68 ab	2.57 ab	2.6 ab
	2.6	2.83 a	2.93 a	2.58 ab	2.53 ab	2.7 ab
Std error = 0.0810						

^aMeans with different letters are significantly different ($p<0.05$).

^bFrankfurter were formulated with 0% (control) or 3% lactate/diacetate solution.

^cNot irradiated (control) or irradiated to 1.8 or 2.6 kGy.

Table 12. Least squares means of sourness for irradiated frankfurters formulated with or without lactate/diacetate and aerobically stored at 4 °C.^a

Ingredient ^b	Irradiation ^c	Wk				
		0	1	2	3	4
Control	Control	2.27 b	2.42 a	2.22 b	2.47 a	1.97 b
	1.8	2.29 b	2.51 a	2.14 b	2.27 b	2.37 ab
	2.6	2.17 b	2.55 a	2.30 ab	2.27 b	2.37 ab
Lactate	Control	2.42 a	2.75 a	2.41 ab	2.63 a	2.80 a
	1.8	2.42 a	2.60 a	2.55 a	2.63 a	2.57 a
	2.6	2.51 a	2.79 a	2.44 a	2.53 a	2.67 a
Std error = 0.0887						

^aMeans with different letters are significantly different ($p<0.05$).

^bFrankfurters were formulated with 0% (control) or 3% lactate/diacetate solution.

^cNot irradiated (control) or irradiated to 1.8 or 2.6 kGy.

Table 13. Least squares means of bitterness for irradiated frankfurters formulated with or without lactate/diacetate and aerobically stored at 4 °C.^a

Ingredient ^b	Irradiation ^c	Wk				
		0	1	2	3	4
Control	Control	2.25 ab	2.23 ab	2.15 ab	2.13 ab	1.47 c
	1.8	2.14 ab	2.21 ab	2.12 ab	2.17 ab	2.2 ab
	2.6	1.97 b	2.14 ab	2.09 ab	2.20 ab	2.13 ab
Lactate	Control	2.28 ab	2.39 a	2.37 ab	2.3 ab	2.37 ab
	1.8	2.34 ab	2.27 ab	2.31 ab	2.33 ab	2.27 ab
	2.6	2.41 a	2.41 ab	2.24 ab	2.20 ab	2.27 ab
Stnd error = 0.0759						

^aMeans with different letters are significantly different ($p < 0.05$).

^bFrankfurters were formulated with 0% (control) or 3% lactate/diacetate solution.

^cNot irradiated (control) or irradiated to 1.8 or 2.6 kGy.

Table 14. Least squares means of metallic flavor and saltiness for irradiated frankfurters formulated with or without lactate/diacetate and aerobically stored at 4 °C for 4 wk.^a

Ingredient ^b	Metallic	Saltiness
Control	2.13 a	6.08 a
Lactate	2.28 b	7.64 b
Stnd error	0.0213	0.1001

^aMeans within a sensory parameter category with different letters are significantly different ($p < 0.05$).

^bFrankfurters were formulated with 0% (control) or 3% lactate/diacetate solution.

Table 15. Least squares means of metallic aftertaste for irradiated frankfurters formulated with or without lactate/diacetate and aerobically stored at 4 °C.^a

Wk	Metallic
0	2.29 a
1	2.24 a
2	2.22 ab
3	2.14 b
4	2.13 b
Stnd error	0.0337

^aMeans with different letters are significantly different ($p < 0.05$).

lactate can have a slight metallic characteristic that may be responsible for this difference.

Table 16 shows a replication by week interaction for the aromatic smoke and feeling factor mouthburn. For both of these attributes, scores were higher for replication 1 than for replications 2 and 3. Both of these attributes are associated with smoke flavor and differences between replications by week may be accounted for by inconsistent application of natural smoke during processing. Smoke flavor of all 3 replications was inconsistent between storage week ranging from 1.65 to 0.95. Mouthburn in replication 1 decreased from 1.39 initially to 0.62 by wk 4, while replications 2 and 3 were less variable ranging from 1.13 to 0.68.

Sweetness and fat mouthfeel aftertaste also varied according to storage week by replication (Table 17). Sweetness in replication 1 was higher than in replications 2 and 3. For replication 1, there was a gradual decline in sweetness score over storage from 1.18 at to 0.62 by wk 4. This loss of sweet taste was not evident in replications 2 and 3 because the attribute was barely perceptible, but consistent. Fat mouthfeel aftertaste scores for replication 1 (2.32 to 2.00) were somewhat higher than those for replications 2 and 3 (1.98 to 1.68), which may be related to meat raw materials differences (Table 18). There were also significant replication by lactate/diacetate interactions for the feeling factor mouthburn and the aftertaste fat mouthfeel. Table 18 shows that replication 3 had a slightly higher mouthburn score (1.14) for lactate/diacetate frankfurters than those without. This difference was not detected for replications 1 or 2. Fat mouthfeel aftertaste

Table 16. Least squares means of smoke flavor and mouthburn feeling factor for irradiated frankfurters formulated with or without lactate/diacetate and aerobically stored at 4 °C.^a

Wk	Smoke			Mouthburn		
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
0	1.60 a	1.15 bc	1.13 bc	1.39 a	0.95 bc	0.93 bc
1	1.65 a	1.10 bc	1.58 a	1.43 a	0.83 bc	1.13 b
2	1.35 bc	1.43 ab	1.30 bc	0.99 bc	1.05 bc	0.80 bc
3	1.85 a	1.07 c	1.53 a	0.82 bc	0.92 bc	0.98 bc
4	1.48 a	0.97 c	0.95 c	0.62 c	0.68 bc	0.92 bc
Std error	0.0786			0.0920		

^aMeans within a sensory parameter category with different letters are significantly different ($p < 0.05$).

Table 17. Least squares means of sweet basic taste and fat mouthfeel for irradiated frankfurters formulated with or without lactate/diacetate and aerobically stored at 4 °C.^a

Wk	Sweetness			Fat mouthfeel		
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
0	1.18 a	0.65 bcd	0.31 de	2.28 a	1.83 b	1.68 b
1	1.06 a	0.43 cde	0.58 cd	2.32 a	1.83 b	1.77 b
2	0.83 b	0.33 de	0.50 cde	2.15 a	1.83 b	1.82 b
3	0.72 bc	0.48 cde	0.22 e	2.00 ab	1.87 b	1.98 b
4	0.62 cd	0.33 de	0.37 de	2.15 a	1.88 b	1.73 b
Std error	0.0713			0.0576		

^aMeans within a sensory parameter category with different letters are significantly different ($p < 0.05$).

Table 18. Least squares means of mouthburn and fat mouthfeel feeling factors for irradiated frankfurters formulated with or without lactate/diacetate and aerobically stored at 4 °C for 4 wk.^a

Ingredient ^b	Mouthburn			Fat mouthfeel		
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
Control	1.00 ab	0.87 ab	0.76 b	2.12 b	1.93 c	1.77 d
Lactate	1.10 a	0.91 ab	1.14 a	2.23 a	1.77 d	1.82 d
Std error	0.0582			0.0364		

^aMeans within a sensory parameter category with different letters are significantly different ($p < 0.05$).

^bFrankfurters were formulated with 0% (control) or 3% lactate/diacetate solution.

was higher for replication 1 than for either replication 2 or 3, which again may be due to inconsistent smoke application or raw material.

Sensory Texture

Overall, both lactate/diacetate addition and irradiation treatments were effective for maintaining sensory texture and slowing product deterioration. Panelist identified differences in both springiness and cohesiveness of mass scores for frankfurters. For both of these attributes, there were significant decreases in the sensory score on wk 4 of storage. Springiness scores for non-irradiated frankfurters without lactate/diacetate averaged 6.65 at wk 4 and was slightly lower than irradiated frankfurters with or without lactate (7.45 to 7.87) (Table 19). In general, no treatment differences were noted for springiness except at wk 4 of storage. Like springiness, cohesiveness of mass scores for the control decreased slightly at wk 4 of storage when compared to all other treatments (Table 20). All other cohesiveness of mass scores across all treatments were similar through 4 wk of storage. Loss of both springiness and cohesiveness late in storage are likely associated with spoilage and product deterioration. Lactate/diacetate and irradiation treatments appeared to have a protective effect and helped retain textural attributes. Perceived juiciness was sustained during storage by the inclusion of lactate/diacetate (Table 21), but a slight decline was noted in frankfurters formulated without lactate/diacetate at 4 wk, when compared to those with lactate/diacetate.

Several replication interactions were identified for sensory texture. As seen in Table 22, replications 2 and 3 were judged to be harder than replication 1. For replications 2 and 3, lactate/diacetate frankfurters were perceived to be slightly harder by

Table 19. Least squares means of springiness for irradiated frankfurters formulated with or without lactate/diacetate and aerobically stored at 4 °C.^a

Ingredient ^b	Irradiation ^c	Wk				
		0	1	2	3	4
Control	Control	7.68 a	7.55 a	7.66 a	7.52 a	6.65 b
	1.8	7.41 a	7.65 a	7.61 a	7.52 a	7.45 a
	2.6	7.37 a	7.57 a	7.52 a	7.38 a	7.50 a
Lactate	Control	7.74 a	8.01 a	7.82 a	7.5 a	7.87 a
	1.8	7.76 a	7.75 a	7.93 a	7.73 a	7.85 a
	2.6	7.94 a	7.91 a	7.83 a	7.82 a	7.68 a

Std error = 0.1146

^aMeans with different letters are significantly different ($p < 0.05$).

^bFrankfurters were formulated with 0% (control) or 3% lactate/diacetate solution.

^cNot irradiated (control) or irradiated to 1.8 or 2.6 kGy.

Table 20. Least squares means of cohesiveness of mass for irradiated frankfurters formulated with or without lactate/diacetate and aerobically stored at 4 °C.^a

Ingredient ^b	Irradiation ^c	Wk				
		0	1	2	3	4
Control	Control	7.42 a	7.51 a	7.55 a	7.33 a	6.68 b
	1.8	7.51 a	7.43 a	7.50 a	7.38 a	7.38 a
	2.6	7.44 a	7.40 a	7.50 a	7.37 a	7.33 a
Lactate	Control	7.42 a	7.55 a	7.52 a	7.15 a	7.47 a
	1.8	7.50 a	7.46 a	7.57 a	7.47 a	7.48 a
	2.6	7.53 a	7.45 a	7.50 a	7.35 a	7.40 a

Std error = 0.0979

^aMeans with different letters are significantly different ($p < 0.05$).

^bFrankfurters were formulated with 0% (control) or 3% lactate/diacetate solution.

^cNot irradiated (control) or irradiated to 1.8 or 2.6 kGy.

Table 21. Least squares means of juiciness for irradiated frankfurters formulated with or without lactate/diacetate and aerobically stored aerobically at 4 °C.^a

Wk	Control ^b	Lactate ^b
0	3.80 a	3.75 a
1	3.68 a	3.54 a
2	3.58 a	3.59 a
3	3.80 a	3.58 a
4	3.30 b	3.51 a
Std error = 0.0575		

^aMeans with different letters are significantly different ($p < 0.05$).

^bFrankfurter were formulated with 0% (control) or 3% lactate/diacetate solution.

Table 22. Least squares means of hardness and juiciness for irradiated frankfurters formulated with or without lactate/diacetate and stored aerobically packaged at 4 °C for 4 wk.^a

Ingredient ^b	Hardness			Juiciness		
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
Control	4.30 c	4.51 b	4.56 b	4.06 b	3.33 d	3.50 c
Lactate	4.38 c	4.72 a	4.74 a	4.26 a	3.17 e	3.35 d
Std error	0.0439			0.0445		

^aMeans within a sensory parameter category with different letters are significantly different ($p < 0.05$).

^bFrankfurters were formulated with 0% (control) or 3% lactate/diacetate solution.

panelists than those without lactate/diacetate. Hardness values actually varied only 0.44 unit between the highest and lowest value. Frankfurters were judged in replication 1 to be juicier than those for replications 2 and 3. Hardness and juiciness scores during storage also varied by replication (Table 23). Trends for replication differences were generally inconsistent and probably due to small differences in processing parameters or raw material variation. Specifically, initial differences in hardness were evident, but did not persist throughout storage. Replications 2 and 3 had an initial score of about 4.9 in contrast to replication 1, which had a score of 4.1. Replications 2 and 3 had slightly higher hardness scores than replication 1 throughout storage.

Juiciness scores for replication 1 were slightly higher than replications 2 and 3 during wk 0 and 1 of storage. By wk 2, replication 1 had declined in juiciness and remained at that level throughout wks 2 through 4 of storage. Replications 2 and 3 tended to have lower juiciness scores through the remainder of storage. Depending upon replication, hardness and juiciness scores for frankfurters varied with small influences from lactate/diacetate.

Sensory Color

Determinations of exterior and interior sensory color were made by the highly trained sensory panel using a predetermined color scale. For all color scores, higher values were darker and lower values were lighter. Most exterior color scores ranged from 2 (peachy tan) to 4 (terra cotta orange) and interior color scores ranged from 2 (fleshy pink) to 4 (apricot). All sensory color differences detected by trained panelists were included in the replication interactions. It is believed that color differences between

Table 23. Least squares means of hardness and juiciness for irradiated frankfurters formulated with or without lactate/diacetate and aerobically stored at 4 °C.^a

Wk	Hardness			Juiciness		
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
0	4.13 a	4.89 c	4.89 c	4.85 a	3.17 d	3.32 d
1	4.65 bc	4.73 bc	4.54 bc	4.38 b	3.15 d	3.31 d
2	4.50 b	4.48 b	4.56 bc	3.92 c	3.18 d	3.65 c
3	4.26 a	4.46 b	4.72 bc	3.93 c	3.60 c	3.53 c
4	4.37 ab	4.53 bc	4.55 bc	3.73 c	3.17 d	3.32 d
Std error	0.0694			0.0704		

^aMeans within a sensory parameter category with different letters are significantly different ($p < 0.05$).

replications were likely due to a malfunction of the smokehouse during the frankfurter cooking cycle, which would explain the inconsistent surface color observed in the frankfurters. As shown in Table 24, irradiation had little effect on exterior color scores. Table 25 shows differences in interior color by storage week and replication. Color scores for replication 1 at 0 and 1 wk were significantly higher (4.29 to 4.33) than those for replications 2 and 3. This may indicate a slight lightening of frankfurter color, however, it was barely perceived by the sensory panel.

The aerobic storage of frankfurters has not been studied as extensively as vacuum packaged product, but sensory changes are expected to occur rapidly after a package is opened. The deterioration processes associated with spoilage can occur to a greater degree in an aerobic system as compared to a vacuum packaged system even with the inclusion of antimicrobial ingredients. It is thought that the sensory changes of aerobically stored frankfurters are associated with microbial growth and potentially the action of bacterial proteolytic enzymes that influence all aspects of sensory analysis. In general, the sensory changes that occurred to the untreated control happened more rapidly than for frankfurters with either or both treatments. Both irradiation and use of lactate/diacetate in a formulation will alter the microbial flora compared to the untreated control. Investigations into this microbial shift and the relative proteolytic activity of each should be further investigated to better understand these changes.

Phase 2

Phase 2 investigated the sensory properties of irradiated frankfurters formulated with or without lactate/diacetate and monitored throughout vacuum packaged storage.

Table 24. Least squares means of external sensory color scores for replication by irradiation interactions for electron beam irradiated frankfurters formulated with or without lactate/diacetate and stored aerobically packaged at 4 °C for 4 wk.^a

Irradiation ^b	Rep 1	Rep 2	Rep 3
Control	3.71 b	3.35 ab	3.20 a
1.8	3.51 b	3.34 ab	3.32 ab
2.6	3.63 b	3.40 ab	3.33 ab
Std error	= 0.0440		

^aMeans with different letters are significantly different ($p<0.05$).

^bNot irradiated (control) or irradiated to 1.8 or 2.6 kGy.

Table 25. Least squares means of internal sensory color scores for irradiated frankfurters formulated with or without lactate/diacetate and stored aerobically packaged at 4 °C.^a

Wk	Rep 1	Rep 2	Rep 3
0	4.29 ab	3.50 c	3.88 b
1	4.33 ab	3.40 c	3.16 c
2	4.38 ab	3.88 b	3.93 b
3	4.28 ab	4.02 ab	3.85 b
4	4.46 a	4.08 ab	4.15 ab
Std error	= 0.1111		

^aMeans with different letters are significantly different ($p<0.05$).

Sensory Aroma

Panelists identified significant aromatics including meaty/brothy complex, fat complex, smoke, and spice complex. As in phase 1, these aroma attributes were expected for frankfurters (Chevance and Farmer 1999). Panelists looked for, but were not able to identify, beefy/brothy, porky/brothy, chicken/brothy, beef fat, pork fat, chicken fat, cardboard, painty, fishy, livery, caramelized, soured, soapy, and wet dog aromatics in significant amounts.

Frankfurter aroma was minimally influenced by the experimental treatments. As shown in Tables 26 and 27, there were replication by week interactions for meaty/brothy, fat complex, smoke, and spice complex flavors. For each of these attributes, mean scores were generally within a narrow range and detected differences were small, inconsistent and presented no clear trend. It is unclear whether or not these replication effects were important in the overall flavor profile of the frankfurter treatments, however, most values were within the expected range for each attribute. Both meaty/brothy complex and fat complex replication interactions may be due to naturally occurring biological variations in meat raw materials such as flavor intensity and degree of lipid oxidation. There were also replication by week interactions for smoke and spice aroma (Table 27). Again, values varied significantly between replications and within each replication but with no clear trends. As previously described, it is thought that the replication interactions for smoke scores may be due to a malfunctioning natural smoke generator. It is unclear why there were differences in spice

Table 26. Least squares means of meaty/brothy complex and fat complex aroma for irradiated frankfurters formulated with or without lactate and stored vacuum packaged at 4 °C.^a

Wk	Meaty/brothy complex			Fat complex		
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
0	2.52 a	2.68 a	2.47 a	0.48 a	0.63 a	0.13 b
2	2.55 a	2.38 a	1.91 b	0.42 ab	0.33 ab	0.17 b
4	2.58 a	2.63 a	2.01 b	0.56 a	0.45 ab	0.35 ab
6	2.29 a	2.23 ab	2.35 a	0.52 a	0.47 a	0.23 b
8	2.58 a	2.43 a	2.65 a	0.13 b	0.28 b	0.22 b
Std error	0.0843			0.0585		

^aMeans within a sensory parameter category with different letters are significantly different ($p < 0.05$).

Table 27. Least squares means of smoke and spice complex aroma for irradiated frankfurters formulated with or without lactate and stored vacuum packaged at 4 °C.^a

Wk	Smoke			Spice complex		
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
0	0.93 b	1.27 a	0.27 c	1.17 b	1.40 ab	1.65 ab
2	1.15 a	0.83 b	0.43 c	1.63 ab	1.75 a	1.62 ab
4	0.15 c	0.30 c	0.69 b	1.63 ab	1.35 ab	1.61 ab
6	0.01 c	0.42 c	0.71 b	1.29 b	1.27 b	1.77 a
8	0.75 b	0.77 b	0.35 c	1.18 b	1.18 b	1.58 ab
Std error	0.0554			0.0967		

^aMeans within a sensory parameter category with different letters are significantly different ($p < 0.05$).

complex aroma. Most mean ranges between replications were relatively small even though detectable by the panel.

Sensory Flavor

Panelists identified attributes important to frankfurter flavor in significant amounts including aromatics (meaty/brothy complex, fat complex, smoke, and spice complex), feeling factors (metallic, astringent, and mouthburn), tastes (salt, sour, bitter, and sweet), and aftertastes (fat mouthfeel and smoke/spice complex). These attributes were expected to be present in frankfurters (Chevance and Farmer 1999). As with phase 1, panelists looked for, but were not able to identify, the aromatics beefy/brothy, porky/brothy, chicken/brothy, beef fat, pork fat, chicken fat, cardboard, painty, fishy, livery, caramelized, soured, soapy, and wet dog.

Overall, most flavor attributes were not strongly influenced by the experimental treatments and were similar to the control in most instances. Table 28 shows a slight loss of smoke flavor during storage for all treatments. These values were low, and may be difficult for consumers to quantify at these low intensity levels. Table 29 shows differences between lactate/diacetate and irradiation treatments for smoke and mouthburn. While significant, these values were low with a narrow range of 0.94 to 1.15 and 0.55 to 0.95, respectively. Smoke flavor and smoky/spicy complex declined in all frankfurters over the storage period (Table 30).

As seen in Tables 31 and 32, there were some replication differences for meaty/brothy complex and bitterness. While detectable by the sensory panel, these

Table 28. Least squares means of smoke flavor for irradiated frankfurters formulated with or without lactate/diacetate and stored vacuum packaged at 4 °C.^a

Wk	Control ^b	Lactate ^b
0	1.18 b	1.47 a
2	1.21 b	1.29 b
4	0.75 cd	0.70 d
6	0.72 d	0.85 cd
8	0.97 c	1.06 bc
Std error = 0.0424		

^aMeans with different letters are significantly different ($p < 0.05$).

^bFrankfurters were formulated with 0% (control) or 3% lactate/diacetate solution.

Table 29. Least squares means of smoke and mouthburn for irradiated frankfurters formulated with or without lactate/diacetate and stored vacuum packaged at 4 °C for 8 wk.^a

Irradiation ^c	Smoke		Mouthburn	
	Control ^b	Lactate ^b	Control ^b	Lactate ^b
Control	0.94 b	1.15 a	0.83 a	0.85 a
1.8	0.95 b	1.06 ab	0.55 b	0.95 a
2.6	1.01 ab	1.01 ab	0.75 a	0.95 a
Std error	0.0329		0.0934	

^aMeans within a sensory parameter category with different letters are significantly different ($p < 0.05$).

^bFrankfurters were formulated with 0% (control) or 3% lactate/diacetate solution.

^cNot irradiated (control) or irradiated to 1.8 or 2.6 kGy.

Table 30. Least squares means of smoke and smoky/spicy complex for irradiated frankfurters formulated with or without lactate/diacetate and stored vacuum packaged at 4 °C.^a

Wk	Smoke			Smoky/spicy		
	Control ^b	1.8 ^b	2.6 ^b	Control ^b	1.8 ^b	2.6 ^b
0	1.27 b	1.47 a	1.23 b	2.18 a	2.20 a	2.25 a
2	1.30 b	1.18 bc	1.27 b	2.13 a	2.16 a	2.18 a
4	0.75 cd	0.67 d	0.75 cd	2.03 a	2.10 a	1.96 a
6	0.87 cd	0.75 cd	0.74 cd	2.24 a	1.89 b	2.10 a
8	1.03 bc	0.97 c	1.03 bc	2.17 a	2.07 a	2.22 a
Std error	0.052			0.0639		

^aMeans within a sensory parameter category with different letters are significantly different ($p < 0.05$).

^bNot irradiated (control) or irradiated to 1.8 or 2.6 kGy.

Table 31. Least squares means of meaty/brothy complex for irradiated frankfurters formulated with or without lactate/diacetate and stored vacuum packaged at 4 °C for 8 wk.^a

Ingredient ^b	Rep 1	Rep 2	Rep 3
Control	4.32 ab	4.40 ab	4.29 ab
Lactate	4.20 b	4.30 ab	4.52 a
Std error	= 0.0660		

^aMeans with different letters are significantly different ($p < 0.05$).

^bFrankfurters were formulated with 0% (control) or 3% lactate/diacetate solution.

Table 32. Least squares means of bitterness for irradiated frankfurters formulated with or without lactate/diacetate and stored vacuum packaged at 4 °C for 8 wk.^a

Ingredient ^b	Rep 1	Rep 2	Rep 3
Control	2.14 b	2.12 b	2.25 a
Lactate	2.38 a	2.33 a	2.31 a
Std error	= 0.0330		

^aMeans with different letters are significantly different ($p < 0.05$).

^bFrankfurters were formulated with 0% (control) or 3% lactate/diacetate solution.

differences may not have an impact on the overall flavor of the frankfurters since the score ranges were small, 4.20 to 4.52 and 2.12 to 2.38, respectively.

Sensory Texture

Table 33 shows small, but significant replication effect for juiciness score differences for frankfurters formulated with or without lactate/diacetate. For replication 1, frankfurters formulated with lactate/diacetate (3.64) were slightly juicier than those without lactate/diacetate (3.48). Over all, juiciness differences were small, but significant.

Springiness differences are presented in Table 34. Replications 1 and 2 had significantly higher springiness scores than replication 3, which had a significantly higher score for frankfurters formulated with lactate/diacetate (7.11) compared to those without lactate/diacetate (6.44). In Table 34, replications 1 and 2 had significantly higher springiness scores (7.68 to 7.51) than replication 3 (6.93 to 6.57). For replication 3, both control and 1.8 kGy treatments had higher springiness scores (6.93 to 6.83) than the 2.6 kGy treatment (6.57). This difference is small but significant and may be due to polymerization of solubilized protein components by irradiation. Overall, both lactate/diacetate and irradiation treatment only had a slight influence on springiness, while lactate/diacetate had a small influence on juiciness. The overall influence of the treatments on texture was not dramatic. However, it is evident that deterioration was minimal and that quality was maintained in contrast to the reductions in quality observed in aerobically packaged frankfurters.

Table 33. Least squares means of juiciness for irradiated frankfurters formulated with or without lactate/diacetate and stored vacuum packaged at 4 °C for 8 wk.^a

Ingredient ^b	Juiciness			Springiness		
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
Control	3.48 b	3.59 ab	3.58 ab	7.49 a	7.58 a	6.44 c
Lactate	3.64 a	3.62 a	3.34 b	7.67 a	7.66 a	7.11 b
Std error	0.0476		0.0521			

^aMeans within a sensory parameter category with different letters are significantly different ($p < 0.05$).

^bFrankfurters were formulated with 0% (control) or 3% lactate/diacetate solution.

Table 34. Least squares means of springiness for irradiated frankfurters formulated with or without lactate/diacetate and stored vacuum packaged at 4 °C for 8 wk.^a

Irradiation ^b	Rep 1	Rep 2	Rep 3
Control	7.68 a	7.56 a	6.93 b
1.8	7.51 a	7.68 a	6.83 b
2.6	7.54 a	7.63 a	6.57 c
Std error	= 0.0638		

^aMeans with different letters are significantly different ($p < 0.05$).

^bNot irradiated (control) or irradiated to 1.8 or 2.6 kGy.

Table 35. Least squares means of external sensory color scores for irradiated frankfurters formulated with or without lactate/diacetate and stored vacuum packaged at 4 °C.^a

Wk	Rep 1	Rep 2	Rep 3
0	3.13 c	2.92 cd	3.60 a
2	2.92 cd	2.83 d	3.65 a
4	2.90 cd	2.73 d	3.55 a
6	3.04 c	2.93 cd	3.40 b
8	3.02 c	2.98 c	3.07 c
Std error	=0.0469		

^aMeans with different letters are significantly different ($p < 0.05$).

Sensory Color

As in phase 1, all sensory color differences detected by the trained panel involved replication interactions. Table 35 shows that exterior sensory color varied by replication during storage. In general, most color scores for replication 3 except wk 8, were significantly darker (3.6 to 3.4) than those for replications 1 and 2 (2.7 to 3.0). Table 36 shows that replication 3 had significantly darker internal color scores (4.08 to 4.16) throughout storage compared to replications 1 and 2 (2.27 to 3.13). Although color score trends varied between replication, the color scores of frankfurters remained fairly consistent throughout storage. As with phase 1, differences between replication for exterior color may be due to the previously mentioned smokehouse malfunctions.

The influence of the experimental treatments on the sensory properties of frankfurters was relatively small compared to those observed for aerobic storage in phase 1. Other authors who have investigated the influence of lactate addition found minimal sensory changes during storage. Nunez and others (2004b) observed small but significant increases in astringency, and bitterness in frankfurters formulated with potassium lactate, however, the overall flavor profile was nearly identical to frankfurters without lactate addition. Terrell and others (1981b and 1982) observed significant sensory changes associated with frankfurters irradiated up to 3.0 kGy, however, the differences in their studies were not quantified by a descriptive attribute panel. It is thought that the sensory changes of vacuum packaged frankfurters may be related to a shift in the microbial population associated with irradiation and use of lactate/diacetate in a formulation. Even though sensory changes of vacuum packaged frankfurters were

Table 36. Least squares means of internal sensory color scores for irradiated frankfurters formulated with or without lactate/diacetate and stored vacuum packaged at 4 °C.^a

Wk	Rep 1	Rep 2	Rep 3
0	2.80 c	2.75 c	4.12 a
2	2.82 c	2.27 e	4.13 a
4	2.54 d	2.37 de	4.16 a
6	3.13 b	2.52 de	4.08 a
8	2.65 cd	2.83 c	4.10 a
Std error = 0.0572			

^aMeans with different letters are significantly different ($p < 0.05$).

much less dramatic than with aerobic frankfurters, the microbial flora would have been altered by the experimental treatments compared to the untreated control. In the vacuum packaged system, even for the untreated control, the flora would have favored lactic acid bacteria instead of aerobic proteolytic bacteria as in phase 1. Again, investigations into this microbial shift and the relative proteolytic activity of each should be further investigated to better understand these changes.

CHAPTER VI

CHEMICAL AND PHYSICAL RESULTS AND DISCUSSION

Phase 1

During phase 1, the chemical and physical properties of irradiated frankfurters formulated with or without lactate/diacetate were monitored throughout aerobic storage.

pH

The pH values of frankfurters varied mainly by formulation but also slightly by irradiation treatment. As expected, frankfurters formulated without lactate/diacetate had significantly higher average pH value (6.50) compared to that with lactate/diacetate (6.31). Table 37 shows non-irradiated frankfurters initially having a pH of 6.40, which declined steadily to about 6.35 at 4 wk. pH values for both irradiation treatments were relatively consistent during storage (6.37 to 6.39).

Lipid Oxidation

Both interactions for TBARS values in phase 1 were related to replication. As shown in Table 38, there was a replication by lactate/diacetate interaction where replication 1 had higher TBARS values (4.5 to 3.52) than both replications 2 and 3 (0.42 to 0.27). Also in replication 1, frankfurters formulated without lactate had higher TBARS values (4.5) than frankfurters formulated with lactate (3.52). Table 39 shows a replication by week interaction for TBARS values. Again, replication 1 had much higher TBARS values (4.87 to 1.41) than replications 2 and 3 throughout storage (0.47 to 0.25). Also, replication 1 had a significantly lower TBARS value (1.41) at wk 1 of storage as compared to subsequent weeks, which were all consistently high. TBARS values that are

Table 37. Least squares means of pH for irradiated frankfurters formulated with or without lactate/diacetate and aerobically stored at 4 °C.^a

Wk	Irradiation ^b		
	0	1.8	2.6
0	6.40 a	6.38 ab	6.38 ab
1	6.38 ab	6.39 ab	6.38 ab
2	6.36 ab	6.37 ab	6.37 ab
3	6.35 b	6.38 ab	6.38 ab
4	6.35 b	6.39 ab	6.39 ab

Std error = 0.0081

^aMeans with different letters are significantly different ($p < 0.05$).

^bNon-irradiated (control) or irradiated to 1.8 or 2.6 kGy.

Table 38. Least squares means of TBARS values for irradiated frankfurters formulated with or without lactate/diacetate and aerobically stored at 4 °C for 4 wk.^a

Ingredient ^b	Rep 1	Rep 2	Rep 3
Control	4.50 a	0.42 c	0.29 c
Lactate	3.52 b	0.39 c	0.27 c

Std error = 0.1253

^aMeans with different letters are significantly different ($p < 0.05$).

^bFrankfurters were formulated with 0% (control) or 3% lactate/diacetate solution.

Table 39. Least squares means of TBARS values of irradiated frankfurters formulated with or without lactate/diacetate and aerobically stored at 4 °C.^a

Wk	Rep 1	Rep 2	Rep 3
0	1.41 b	0.47 c	0.25 c
1	4.84 a	0.41 c	0.27 c
2	4.60 a	0.46 c	0.28 c
3	4.32 a	0.31 c	0.32 c
4	4.87 a	0.39 c	0.27 c

Std error = 0.1981

^aMeans with different letters are significantly different ($p < 0.05$).

this high, especially in the initial stages of storage are likely due to low quality raw materials. Although fresh mechanically deboned poultry was ordered, there was no assessment at delivery regarding the quality characteristics of the poultry component. It is likely that the mechanically deboned poultry used for replication 1 was compromised, which would explain the high TBARS values. Mechanically deboned poultry can deteriorate and oxidize quickly even at frozen temperatures. Irradiation treatments did not appear to increase TBARS values of frankfurters for any of the treatments.

Instrumental Color

Both exterior and interior color values were determined for frankfurters. Exterior L* values varied only slightly between sampling wk 2 and 3, while exterior b* values became more yellow during wk 3 and 4 of storage (Table 40). Including lactate/diacetate in a frankfurter formulation increased the initial redness of frankfurters (17.14 in contrast to 15.62), but all exterior a* values for frankfurters with and without lactate/diacetate declined during storage (Table 41). Exterior a* values were not different between irradiation treatments except at wk 6 and 8 where redness loss was greater for irradiated frankfurters than non-irradiated frankfurters (Table 42). Exterior a* values for irradiation treatments declined or became less red during storage, and 2.6 kGy irradiated red color faded more than non-irradiated and 1.8 kGy frankfurters. Overall, lactate/diacetate addition to formulations made the exterior color of frankfurters slightly redder, but as with irradiation, the redness faded with storage. The exterior a* value for control frankfurters in replication 1 (16.65) was slightly redder than replications 2 and 3 (15.06 and 15.09, respectively) (Table 43), but exterior a* values for replications 2 and 3

Table 40. Least squares means of exterior L* and b* values for irradiated frankfurters formulated with or without lactate/diacetate and aerobically stored at 4 °C.^a

Wk	L*	b*
0	64.4 ab	15.21 a
1	64.08 ab	15.49 a
2	64.63 a	15.40 a
3	63.85 b	16.04 b
4	64.35 ab	16.19 b
Std error	0.1769	0.1659

^aMeans within each color space value with different letters are significantly different ($p<0.05$).

Table 41. Least squares means of exterior a* values for irradiated frankfurters formulated with or without lactate/diacetate and aerobically stored at 4 °C.^a

Wk	Control ^b	Lactate ^b
0	15.62 c	17.14 d
1	15.35 c	17.17 d
2	14.38 b	14.52 b
3	14.31 b	15.70 c
4	13.32 a	14.53 b
Std error	= 0.2527	

^aMeans with different letters are significantly different ($p<0.05$).

^bFrankfurters were formulated with 0% (control) or 3% lactate/diacetate solution.

Table 42. Least squares means of exterior a* values for irradiated frankfurters formulated with or without lactate/diacetate and aerobically stored at 4 °C.^a

Wk	Irradiation ^b		
	0	1.8	2.6
0	16.58 a	16.26 a	16.30 a
1	16.63 a	16.39 a	15.76 a
2	14.74 b	14.35 b	14.27 b
3	15.75 a	14.54 b	14.73 b
4	15.16 ab	13.94 b	12.67 c
Std error	= 0.3094		

^aMeans with different letters are significantly different ($p<0.05$).

^bNot irradiated (control) or irradiated to 1.8 or 2.6 kGy.

Table 43. Least squares means of exterior a^* values for irradiated frankfurters formulated with or without lactate/diacetate and aerobically stored at 4 °C for 4 wk.^a

Ingredient ^b	Rep 1	Rep 2	Rep 3
Control	16.65 a	15.06 c	15.09 c
Lactate	14.27 b	16.59 d	16.58d
Std error = 0.1957			

^aMeans with different letters are significantly different ($p < 0.05$).

^bFrankfurters were formulated with 0% (control) or 3% lactate/diacetate solution.

showed frankfurters with lactate/diacetate to be more red than replication 1. Replication interactions for exterior color may be attributed to a smoke generator malfunction that caused uneven smoke distribution on the frankfurter's surface of replication 1. Based on the a^* values presented in Table 41 and replications 2 and 3 in Table 43, it is believed that the true treatment effects of lactate/diacetate are that they increased exterior a^* values in frankfurters causing a slightly redder surface.

Although there was a small, but significant, decline in interior L^* value on wk 3 in comparison to wk 4, all other lightness values were not different (Table 44). The interior of all frankfurters became slightly more yellow over 8 wk of storage (Tables 45 and 46) and irradiation treatments made the interior of frankfurters more yellow on storage wk 6 and 8. As shown in Table 46, interior a^* or redness values declined as storage progressed. There were several minor replication by week interactions for interior a^* and b^* color (Table 46). Interior L^* values were different among replications with replication 1 having a slightly darker color (66.52) compared to replications 2 and 3 (67.26 and 67.02), respectively (data not shown). In all replications and in most instances, frankfurters lost a small amount of internal redness and became slightly more yellow during storage. Specifically, replication 1 had slightly lower interior a^* values (13.39 to 11.28) than replications 2 and 3 (14.93 to 12.15). In addition, for all storage dates except wk 4, interior b^* values for replication 1 were slightly higher (13.18 to 11.80) compared to those for replications 2 and 3 (12.17 to 11.07). These interior color variations may be due to processing conditions or raw material variations between replications.

Table 44. Least squares means of interior L* values for irradiated frankfurters formulated with or without lactate/diacetate and aerobically stored at 4 °C.^a

Wk	L*
0	67.21 ab
1	66.87 ab
2	66.80 ab
3	66.44 a
4	67.33 b

Std error = 0.1725

^aMeans with different letters are significantly different ($p < 0.05$).

Table 45. Least squares means of internal b* values for irradiated frankfurters formulated with or without lactate/diacetate and aerobically stored at 4 °C.^a

Wk	Irradiation ^b		
	0	1.8	2.6
0	11.38 a	11.34 a	11.31 a
1	11.48 a	11.60 a	11.63 a
2	12.47 c	12.59 c	12.77 c
3	11.67 a	12.06 b	12.16 b
4	12.07 b	12.60 c	12.79 c

Std error = 0.1049

^aMeans with different letters are significantly different ($p < 0.05$).

^bNot irradiated (control) or irradiated to 1.8 or 2.6 kGy.

Table 46. Least squares means of internal a* and b* values for irradiated frankfurters formulated with or without lactate/diacetate and aerobically stored at 4 °C.^a

Wk	a*			b*		
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
0	13.39 bc	14.93 d	14.89 d	11.80 b	11.15 a	11.07 a
1	13.71 c	14.50 d	14.16 d	12.34 cd	11.18 a	11.20 a
2	13.05 bc	13.2 bc	13.11 bc	12.44 cd	12.54 cd	12.85 d
3	12.59 bc	13.86 cd	13.93 cd	12.71 d	11.64 b	11.55 b
4	11.28 a	12.15 b	12.78 bc	13.18 e	12.17 c	12.12 c

Std error

0.1049

^aMeans within each color space value with different letters are significantly different ($p < 0.05$).

As with microbial and sensory analysis, limited work has been done regarding the safety and quality attributes of aerobically stored frankfurters, irradiated or otherwise. Important changes can occur during aerobic storage that may be different than vacuum packaged storage.

Phase 2

Again, for phase 2, the chemical and physical properties of irradiated frankfurters formulated with or without lactate/diacetate were monitored throughout vacuum packaged storage.

pH

A week by lactate/diacetate interaction was present for frankfurter pH. Frankfurters formulated with lactate/diacetate had a significantly lower pH (6.29 to 6.33) than frankfurters without lactate/diacetate (6.47 to 6.53) for all sampling dates (Table 47). Even though significant, all lactate/diacetate treatment differences were small and similar to other studies.

Lipid Oxidation

In phase 2, both lactate/diacetate formulation and irradiation treatments appeared to slightly influence TBARS values (Table 48). Overall, TBARS values for all treatments were in a narrow range (2.18 to 2.40) and minimally influenced by irradiation. The highest TBARS value for all treatments was 2.4 for frankfurters formulated with lactate/diacetate and irradiated to 1.8 kGy. The lowest TBARS value was 2.18 for frankfurters formulated with lactate/diacetate and irradiated to 2.6 kGy.

Table 47. Least squares means of pH for irradiated frankfurters formulated with or without lactate/diacetate and stored vacuum packaged at 4 °C.^a

Ingredient ^b		
Wk	Control	Lactate
0	6.53 a	6.32 c
2	6.48 b	6.31 cd
4	6.47 b	6.33 c
6	6.49 b	6.31 cd
8	6.48 b	6.29 d
Std error = 0.0089		

^aMeans with different letters are significantly different ($p < 0.05$).

^bFrankfurters were formulated with 0% (control) or 3% lactate/diacetate solution.

Table 48. Least squares means of TBARS value for irradiated frankfurters formulated with or without lactate/diacetate and stored vacuum packaged 4 °C for 8 wk.^a

Irradiation ^c			
Ingredient ^b	0	1.8	2.6
Control	2.33 ab	2.27 ab	2.27 ab
Lactate	2.28 ab	2.40 a	2.18 b
Std error = 0.0355			

^aMeans with different letters are significantly different ($p < 0.05$).

^bFrankfurters were formulated with 0% (control) or 3% lactate/diacetate solution.

^cNot irradiated (control) or irradiated to 1.8 or 2.6 kGy.

There was a significant replication by lactate/diacetate interaction (Table 49) showing that replication 3 had nearly double the TBARS values (3.33 to 3.13) of replications 1 or 2 (1.87 to 1.69). This same increase was noted in Table 50, where replication 3 had higher TBARS values (3.70 to 2.89) than both replications 1 and 2 (1.94 to 1.41) at each sampling week. Although TBARS values were higher for replication 3, the trained sensory panel reported no significant off-flavors or aromatics representative of lipid oxidation. Thus, the TBARS results may reflect chemical changes in the frankfurters, but these had no impact on flavor or aroma. As in phase 1, these replication interactions may be attributed to a decline in the quality of mechanically deboned poultry which was stored frozen, but may have had a higher level of oxidization.

Instrumental Color

Overall, few differences were noted for exterior frankfurter color. Table 51 shows b^* values (yellowness) over 8 wk storage. Comparisons of the control and lactate/diacetate treatments by week show few differences in exterior b^* values. Both frankfurters formulated with or without lactate/diacetate became slightly more yellow during storage, increasing exterior b^* values from 15.78 to 15.38 initially to 17.58 to 17.56 at wk 3. After 4 wk storage, frankfurters without lactate decreased in exterior b^* value to 15.84 while frankfurters with lactate/diacetate remained similar to wks 4 and 6 at 16.90. Exterior L^* and a^* values were not influenced by the treatments.

Several replication interactions were identified for exterior color. Table 52 shows a replication by lactate/diacetate interaction for exterior L^* and a^* values. Frankfurters were slightly lighter in replication 3 (63.27 to 61.57) compared to replications 1 and 2

Table 49. Least squares means of TBARS value for irradiated frankfurters formulated with or without lactate/diacetate and stored vacuum packaged at 4 °C for 8 wk.^a

Ingredient ^b	Rep 1	Rep 2	Rep 3
Control	1.74 cd	1.69 d	3.44 a
Lactate	1.87c	1.80 c	3.13 b
Std error = 0.0355			

^aMeans with different letters are significantly different ($p < 0.05$).

^bFrankfurters were formulated with 0% (control) or 3% lactate/diacetate solution.

Table 50. Least squares means of TBARS value for irradiated frankfurters formulated with or without lactate/diacetate and stored vacuum packaged at 4 °C.^a

Wk	Rep 1	Rep2	Rep 3
0	1.92 d	1.87 de	3.70 a
2	1.75 de	1.67 e	3.43 b
4	1.93 d	1.87 de	3.24 b
6	1.94 d	1.92 d	2.89 c
8	1.51 ef	1.41 f	3.28 b
Std error = 0.0562			

^aMeans with different letters are significantly different ($p < 0.05$).

Table 51. Least squares means of exterior b* values for irradiated frankfurters formulated with or without lactate/diacetate and stored vacuum packaged at 4 °C.^a

Wk	Control ^b	Lactate ^b
0	15.38 a	15.78 ab
2	15.76 ab	16.12 b
4	17.33 c	17.36 c
6	17.58 c	17.56 c
8	15.84 ab	16.90 c
Std error = 0.1701		

^aMeans with different letters are significantly different ($p < 0.05$).

^bFrankfurters were formulated with 0% (control) or 3% lactate/diacetate solution.

Table 52. Least squares means of exterior L* and a* values for irradiated frankfurters formulated with or without lactate/diacetate and stored vacuum packaged at 4 °C for 8 wk.^a

Ingredient ^b	L*			a *		
	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
Control	60.42 a	60.00 a	63.27 b	16.89 a	17.26 a	15.06 b
Lactate	60.21 a	59.98 a	61.57 b	16.89 a	17.03 a	16.15 b
Std error	0.1742			0.1612		

^aMeans within each color space value with different letters are significantly different ($p < 0.05$).

^bFrankfurter were formulated with 0% (control) or 3% lactate/diacetate solution.

(60.42 to 59.98). Also, frankfurters from replications 1 and 2 (17.26 to 17.03) were slightly redder than those from replication 3 (16.15 to 15.06). Table 53 shows replication interactions for exterior L^* , a^* , and b^* values. During wk 0 and 2 of storage, frankfurters from replications 1 and 2 (58.11 to 58.58) were slightly darker than those from replication 3 (63.44 to 61.96). A similar situation was noted for exterior a^* and b^* values. Exterior a^* color space values for replication 3 (15.28 to 15.84) were less red than those for replications 1 and 2 (18.52 to 18.81) on wk 0 and 2, after which they decrease and increase slightly on wk 4. Exterior b^* values were higher for replication 3 (16.51 to 17.72) than replications 1 and 2 (14.98 to 15.15) during the first 2 wk of storage, remained constant on wks 4 and 6 and then declined in replication 1 and 2 at wk 8. As stated previously, replication interactions for external color may be due to a smokehouse malfunction that could have produced inconsistent exterior color.

All differences for interior color of phase 2 involved replication interactions. Table 54 shows replication by lactate/diacetate differences for interior L^* , a^* , and b^* values. In general, frankfurters from replication 3 were slightly whiter, less red, and more yellow than those from replications 1 and 2. There were also replication by week interactions for interior L^* , a^* , and b^* values (Table 55). Interior color was viewed as fairly consistent throughout storage with differences that can be attributed to inherent biological variation between replications. These differences may have been due to variations in meat raw materials or slight variations in processing.

A number of studies have been performed using vacuum packaged frankfurters or bologna either formulated with lactate, treated with irradiation, a combination of the

Table 53. Least squares means of exterior L*, a*, and b* values for irradiated frankfurters formulated with or without lactate/diacetate and stored vacuum packaged at 4 °C.^a

	L*			a*			b*		
Wk	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
0	58.13 a	58.11 a	63.44 d	18.69 a	18.81 a	15.28 b	15.15 ab	15.09 ab	16.51 c
2	58.23 a	58.58 a	61.96 c	18.52 a	18.52 a	15.84 b	15.11 ab	14.98 a	17.72 d
4	63.13 cd	62.12 c	62.50 cd	16.00 b	16.48 b	15.96 b	17.16 d	17.65 d	17.22 d
6	61.95 c	61.91 c	61.99 c	15.10 b	15.59 b	15.16 b	17.38 d	17.53 d	17.80 d
8	60.16 b	59.24 a	62.21 c	16.14 b	16.31 b	15.78 b	15.52 ab	15.65 b	17.94 d
Std error	0.2754			0.2560			0.2084		

^aMeans within each color space value with different letters are significantly different ($p<0.05$).

Table 54. Least squares means of internal L*, a*, and b* values for irradiated frankfurters formulated with or without lactate/diacetate and stored vacuum packaged at 4 °C for 8 wk.^a

Int L	L*			a*			b*		
Ingredient ^b	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
Control	64.33 b	63.88 a	66.16 d	15.04 c	15.02 c	13.36 a	12.12 b	11.98 ab	12.95 d
Lactate	64.30 b	64.29 b	65.52 c	14.97 c	14.91 c	13.93 b	11.98 ab	11.93 a	12.49 c
Std error	0.1089			0.0785			0.0663		

^aMeans within each color space value with different letters are significantly different ($p<0.05$).

^bFrankfurters were formulated with 0% (control) or 3% lactate/diacetate solution.

Table 55. Least squares means of internal L*, a*, and b* values for irradiated frankfurters formulated with or without lactate/diacetate and stored vacuum packaged at 4 °C.^a

	L*			a*			b*		
Wk	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3	Rep 1	Rep 2	Rep 3
0	62.61 b	62.48 ab	66.59 f	16.05 d	16.03 d	13.08 a	11.45 b	11.14 a	12.00 b
2	62.17 ab	62.05 a	65.46 e	16.52 d	16.37 d	13.84 bc	11.62 b	11.58 b	12.97 c
4	66.52 f	66.47 f	66.37 f	14.04 bc	14.01 bc	13.57 b	12.97 c	12.65 c	12.69 c
6	66.18 f	66.16 f	65.31 e	14.08 bc	14.13 bc	14.00 bc	12.49 c	12.62 cd	13.08 c
8	64.1 d	63.3 c	65.49 e	14.33 c	14.29 c	13.72 bc	11.73 b	11.79 b	12.88 c
Std error	0.1715			0.1241			0.1048		

^aMeans within each color space value with different letters are significantly different ($p<0.05$).

two or other antimicrobial treatments (Nunez and others 2004b; Sommers and Fan 2002, 2003; Sommers and others 2003a, 2003b). In each case, changes in lipid oxidation and instrumental color were minimal and barely detectable.

CHAPTER VII

SUMMARY AND CONCLUSIONS

The use of potassium lactate and sodium diacetate in frankfurter formulations in combination with pasteurizing doses of irradiation were effective for controlling the growth of *Listeria monocytogenes* on frankfurters as well as maintaining sensory attributes. In an aerobic system where irradiation occurred before inoculation, the incorporation of lactate/diacetate had a listeristatic effect through 4 wk of refrigerated storage. Irradiation did not reduce initial bacterial numbers. Both APC and *L. monocytogenes* counts on frankfurters with lactate/diacetate remained constant while those without lactate/diacetate increased steadily from 5.4 to 9.3 log cfu. All frankfurters formulated with lactate/diacetate effectively suppressed outgrowth of *L. monocytogenes*. Frankfurters without lactate/diacetate and treated only with irradiation had higher *L. monocytogenes* counts after 2 wks. Both treatments together or alone were not detrimental to sensory aroma, flavor or texture attributes. Sensory color was not dramatically influenced by either treatment, however, L^* , b^* and especially a^* values of all treatments declined during storage. Lactate/diacetate addition to frankfurters made them slightly redder, but the redness of all treatments faded during storage. Lactate and diacetate incorporation into a frankfurter formulation was effective for retarding the outgrowth of *L. monocytogenes* aerobically. Use of this antimicrobial blend could enhance the safety of refrigerated products accidentally contaminated after opening or contaminated by a consumer for up to 4 wk while maintaining desirable sensory attributes.

In a vacuum packaged system, exposure of frankfurters to pasteurizing irradiation at 1.8 or 2.6 kGy and the addition of lactate/diacetate to a formulation were effective for controlling the outgrowth of *L. monocytogenes* in an unopened vacuum package. Incremental reductions were observed in both APC and *L. monocytogenes* counts immediately following irradiation treatments of 1.8 and 2.6 kGy. Initial total microbial loads in irradiated frankfurters with or without lactate/diacetate declined by 3 and 5 log cfu/frankfurter, respectively. These reductions were maintained throughout 8 wk vacuum packaged storage for frankfurters with lactate/diacetate addition, however, microbial counts for frankfurters without lactate/diacetate, but irradiated, eventually increased during the last week of storage. There were few treatment effects on flavor, odor, texture or color attributes of vacuum packaged frankfurters as compared to those that were aerobically stored. Instrumental L*a*b* color scores varied slightly, but were fairly consistent throughout storage. Overall, the combination of pasteurizing doses of irradiation and lactate/diacetate were effective for retarding *L. monocytogenes* growth and keeping counts constant throughout vacuum packaged storage. The combination of these two antimicrobial factors could effectively retard the growth of *L. monocytogenes* on frankfurters, and if recontamination did occur after opening, outgrowth would be prevented.

This work has characterized the effectiveness lactate/diacetate incorporated into frankfurters in combination with pasteurizing doses of irradiation for suppressing *L. monocytogenes* growth under both aerobic and anaerobic refrigerated storage conditions. The study was undertaken to understand the complexities of producing a further

processed meat product with the highest level of safety and quality possible, and to provide validation of the processing conditions. Because limited work has been done to evaluate the potential risks that contaminated aerobically stored frankfurters pose, these findings should provide essential information to industry and regulatory agencies charged with protecting public safety.

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APPENDIX A

Frankfurter Processing Specifications

Specifications and Formulas

Fresh and/or frozen mechanically deboned chicken, lean beef trimmings and pork fat trimmings (-2° to 3 °C) were selected, coarse ground through a 1.27 cm (1/2”) plate, reground through a 0.48 cm (3/16”) plate, analyzed for fat content and formulated to achieve a ~28% fat endpoint. A base formulation of raw meat materials, dry ingredients and water are shown in Table 56 and represent calculations to yield a finished product with the following specifications:

- 70 to 75% meat block
- ~28% ether extractable fat
- 56 to 58% moisture
- pH 6.0 to 6.3
- 2.0% salt (sodium chloride)
- 0.35 to 0.5% sodium tripolyphosphate
- <156 mg/kg (ppm) of sodium nitrite (calculated on the meat component)
- 3.0% potassium lactate/sodium diacetate (ingredient treatment only)
- Achieve an endpoint temperature of 71.1 °C (160 °F) as monitored by calibrated thermocouple

Processing Procedures

1. Temper fresh and/or frozen beef lean, poultry and pork fat trimmings to -3.4 to -2.2°C (26-28 °F).
2. Grind tempered beef lean, poultry and fat trimmings through a 1.27 cm (1/2”) plate, regrind separately through 0.48 cm (3/16”) plate or smaller.
3. Combine lean beef trim, 1/2 the total amount of chilled water (slush), pre-blended spices, salt, sodium erythorbate, sodium tripolyphosphate and nitrite in bowl chopper.
4. Mince/chop for 5 minutes keeping the temperature <4.5 °C (40 °F).
5. Rest batter for 1-3 min (salt soluble protein extraction), add fat trim, remaining ice and other ingredients, chop for 3-5 minutes to an endpoint temperature of <15.6 °C (60 °F).
6. Discharge meat batter, vacuum stuff into inedible 28 mm cellulose casings.
7. Thermally process and smoke according to the smokehouse schedule (Table 57) to an end point temperature of 71.1 °C (160 °F).
8. Cold water shower for 6-8 minutes until an internal temperature of <38 °C (100 °F), chill overnight to 4.5 °C (40 °F), peel casings and package as specified.

Table 56. Raw batch weight frankfurter formulation to yield a 28% fat finished product.

Ingredients	Formulation Treatments	
	Control (%)	Lactate/ Diacetate (%)
Meat Trimmings	<u>75.0</u>	<u>75.0</u>
Lean beef trim (85/15)	22.5	22.5
Pork fat trim (60/40)	32.7	32.7
Mechanically Deboned Chicken (17% Fat)	19.8	19.8
Non-meat Ingredients	<u>25.9</u>	<u>25.9</u>
Salt	1.66	1.66
Lactate/ Diacetate (as specified)	-	3.0
Corn Syrup Solids (DE 42)*	1.48	1.48
HMP or HVP	0.74	0.74
Hydrolyzed Beef Stock	0.37	0.37
Sodium Tripolyphosphate	0.33	0.32
Spice/ Seasoning	0.37	0.37
Sodium Erythorbate	0.037	0.037
Sodium Nitrite (cure blend)**	0.185	0.185
Added water	13.3	10.3
10% added water (Cook Shrink)	7.4	7.2
Total (Batter)	100.9	100.9

* DE = Dextrose Equivalent

** Cure blend contains 6.25% sodium nitrite bonded to 93.75% salt. Pure nitrite, if used, would be added at 0.011% while the salt would be increased to 1.84%.

Table 57. Smokehouse schedule – frankfurters (28% fat).

Time (min)	Dry Bulb (F)	Wet Bulb (F)	R.H. (%)	Smoke
30	130°	96°	29	Off
15	140°	104°	30	On
15	150°	112°	30	On
15	160°	120°	31	On
15	170°	128°	31	On
15	180°*	134°	30	On

* Hold to endpoint internal temperature of 71.1 °C (160 ° F); shower to 35 °C (95 °F) then; chill to <4.5 °C (40 °F). See Appendix B, USDA-FSIS for chilling requirements at (<http://www.fsis.usda.gov/oa/fr/95033f%2Da.htm>)

APPENDIX B

Table 58. Frankfurter flavor/texture/aroma profile ballot.

SAMPLE ID #:	W/U	182	182	880	880	102	102
		Aroma		Aroma		Aroma	
AROMATICS:							
Beefy/Brothy							
Porky/Brothy							
Chicken/Brothy							
Meaty/Brothy							
Beef Fat							
Pork Fat							
Chicken Fat							
Fat Complex							
Smoke							
Spice Complex							
Cardboard							
Painty							
Fishy							
Livery							
Caramelized							
Soured							
Soapy							
Wet Dog							
Other (describe)							
FEELING FACTORS:							
Metallic							
Astringent							
Mouthburn							
TASTES:							
Salt							
Sour							
Bitter							
Sweet							
AFTERTASTES:							
Fat Mouthfeel							
Smoke/Spice Complex							
Other (describe)							
TEXTURE							
Springiness							
Hardness							
Juiciness							
Cohesiveness of Mass							

Table 59. Frankfurter color ballot.

Sample Number	External Color	Internal Color
874		
545		
577		
819		
346		
623		
187		
737		
538		
164		
147		
752		

APPENDIX C

Table 60. Sensory attribute descriptions.

Attribute	Description
Aromatics	
Meaty/brothy complex	The brown aromas or flavors associated with broth but not with any particular species.
Fat complex	The aromas and flavor associated with meat fat but not with any particular species.
Smoke	The dark brown aromas and flavors associated with burning or charred wood.
Spice complex	The aromas and flavors associated with spices but not any particular spice.
Feeling factors	
Metallic	The sensations on the tongue associated with metals such as iron or copper.
Astringent	The shrinking or puckering of the tongue surface caused by substances such as tannins or alum.
Mouthburn	The burning sensation of the mouth caused by substances such as capsaicin or piperdine.
Tastes	
Salt	The taste stimulated by sodium salts such as sodium chloride.
Sour	The taste stimulated by acids such as citric or malic acid.
Bitter	The taste stimulated by such substances such as quinine or caffeine.
Sweet	The taste stimulated by sugars such a sucrose or fructose.
Aftertastes	
Fat mouthfeel	The amount of oily film remaining on the oral palate after expectorating.
Smoke/spice complex	The amount of smoke/spice complex aromas or flavors remaining after expectorating.
Texture	
Springiness	The amount a sample returns to original shape after a certain time period.
Hardness	The force required to bite through a sample.
Juiciness	The amount of wetness released from the sample upon biting.
Cohesiveness of mass	The degree to which the sample mass holds together after chewing.

APPENDIX D

Table 61. Least squares means for APC counts (log cfu/frank) of frankfurters treated with electron beam irradiation before inoculation, formulated with or without lactate/diacetate, and aerobically packaged at 4 °C.^a

Ingredient ^b	Irradiation ^c	Wk				
		0	1	2	3	4
Control	Control	5.64 ab	6.55 ab	7.83 c	8.57 cd	9.79 d
	1.8 kGy	5.37 ab	6.20 ab	6.72 b	8.03 c	9.33 cd
	2.6 kGy	5.71 ab	5.84 ab	6.80 b	8.02 c	9.16 cd
Lactate	Control	5.26 ab	5.53 ab	5.25 ab	5.61 ab	6.50 ab
	1.8 kGy	5.10 ab	5.17 ab	5.17 ab	5.03 ab	4.73 a
	2.6 kGy	5.19 ab	5.10 ab	5.18 ab	4.84 ab	4.97 ab
Stnd error		= 0.3518				

^aMeans with different letters are significantly different ($p < 0.05$).

^bFrankfurters were formulated with 0% (control) or 3% lactate/diacetate (lactate).

^cNot irradiated (control) or irradiated to 1.8 or 2.6 kGy.

Table 62. Least squares means for *Listeria monocytogenes* counts (log *L. monocytogenes*/frank) of frankfurters treated with electron beam irradiation before inoculation, formulated with lactate/diacetate, and stored aerobically packaged at 4 °C.^a

Ingredient ^b	Irradiation ^c	Wk				
		0	1	2	3	4
Control	Control	5.45 ab	5.46 ab	5.89 ab	7.85 cd	8.67 d
	1.8 kGy	5.27 ab	6.06 ab	6.99 c	7.80 c	9.33 d
	2.6 kGy	5.47 ab	6.14 ab	7.19 c	8.07 d	9.21 d
Lactate	Control	5.11 a	5.58 ab	6.67 bc	5.82 ab	6.31 ab
	1.8 kGy	5.26 ab	5.39 ab	5.60 ab	5.18 a	4.70 a
	2.6 kGy	5.28 ab	5.23 a	5.52 ab	5.31 ab	5.34 ab
Stnd error		= 0.2924				

^aMeans with different letters are significantly different ($p < 0.05$).

^bFrankfurters were formulated with 0% (control) or 3% lactate/diacetate (lactate).

^cNot irradiated (control) or irradiated to 1.8 or 2.6 kGy.

Table 63. Least squares means for APC counts (log cfu/frank) of frankfurters treated with electron beam irradiation after inoculation, formulated with lactate/diacetate, and stored vacuum packaged at 4 °C.^a

Ingredient ^b	Irradiation ^c	Wk				
		0	2	4	6	8
Control	Control	7.66 a	7.91 a	8.22 a	8.70 a	8.32 a
	1.8 kGy	4.60 b	3.55 bc	5.59 b	6.94 a	7.38 a
	2.6 kGy	2.72 cd	1.14 d	2.51 cd	1.09 d	5.43 b
Lactate	Control	7.52 a	7.47 a	7.45 a	7.27 a	7.27 a
	1.8 kGy	4.12 b	3.65 bc	3.81 b	3.65 bc	3.62 bc
	2.6 kGy	2.03 cd	1.27 d	1.95 cd	1.23 d	1.60 cd
Std error		= 0.4132				

^aMeans with different letters are significantly different (p<0.05).

^bFrankfurters were formulated with 0% (control) or 3% lactate/diacetate (lactate).

^cNot irradiated (control) or irradiated to 1.8 or 2.6 kGy.

Table 64. Least squares means for *Listeria monocytogenes* counts (log *L. monocytogenes*/frank) of frankfurters treated with electron beam irradiation after inoculation, formulated with lactate/diacetate, and stored vacuum packaged at 4 °C.^a

Ingredient ^b	Irradiation ^c	Wk				
		0	2	4	6	8
Control	Control	7.65 a	6.96 a	7.97 a	8.36 a	8.09 a
	1.8 kGy	4.69 bc	3.69 bc	6.28 ab	6.35 ab	7.43 a
	2.6 kGy	2.55 c	1.47 c	2.43 c	1.47 c	6.13 ab
Lactate	Control	7.63 a	7.47 a	7.70 a	7.31 a	7.42 a
	1.8 kGy	4.24 bc	3.75 bc	3.99 bc	3.76 bc	3.00 bc
	2.6 kGy	2.07 c	1.47 c	2.76 c	2.68 c	2.46 c
Std error		= 0.6111				

^aMeans with different letters are significantly different (p<0.05).

^bFrankfurters were formulated with 0% (control) or 3% lactate/diacetate (lactate).

^cNot irradiated (control) or irradiated to 1.8 or 2.6 kGy.

VITA

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